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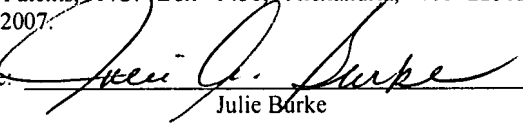
**IN THE UNITED STATES PATENT AND TRADEMARK OFFICE
BEFORE THE BOARD OF PATENT APPEALS AND INTERFERENCES**

Appl. No. : 09/915,150 Confirmation No. 6442
Applicant : Gary R. DelDuca *et al.*
Filed : July 25, 2001
Title : METHODS FOR MAKING MODIFIED ATMOSPHERIC PACKAGES
TC/A.U. : 1761
Examiner : Jyoti Chawla
Docket No. : 247097-001080USPT

TRANSMITTAL OF APPEAL BRIEF

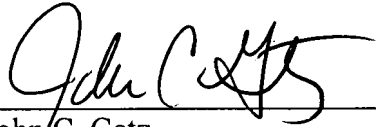
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Signature:	 Julie Burke

Submitted herewith is Appellants' Appeal Brief in support of the Notice of Appeal filed on May 8, 2007. Please charge the amount of \$500.00 for the fees due in connection with the filing of the Notice of Appeal in accord with 37 C.F.R. § 41.20(b)(1), to Deposit Account 50-4181 (Attorney Docket No. 247097-001080USPT) and please credit any additional fees to such deposit account.

Date: May 18, 2007



John C. Gatz
Registration No. 41,774
NIXON PEABODY LLP
161 North Clark Street, Suite 4800
Chicago, Illinois 60601
(312) 425-3900 – Telephone

Attorney for Appellants



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PATENT

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Docket No. : 247097-001080USPT

APPEAL BRIEF PURSUANT TO 37 C.F.R. § 41.37

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Signature: _____

Julie Burke

Dear Commissioner:

This appeal brief is filed pursuant to Appellants' appeal to the Board of Patent Appeals and Interferences from the final rejection of claims 1-37, 87-90 and 161-171 in an Office Action dated February 8, 2007, for the above-listed application.

1. REAL PARTY IN INTEREST

The real party in interest is Pactiv Corporation, a corporation organized and existing under the laws of the State of Delaware, having its principal place of business at 1900 West Field Court, Lake Forest, IL 60045.

2. RELATED APPEALS AND INTERFERENCES

This appeal is related to the appeal filed in Application No. 10/190,375. The Notice of Appeal was filed on February 6, 2007 and the corresponding appeal brief was filed on April 6, 2007. There are no other related appeals and interferences.

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3. STATUS OF CLAIMS

Claims 1-37, 87-90 and 161-171 are pending and have been finally rejected. Claims 38-86 and 91-160 were previously cancelled. It is from the final rejection of claims 1-37, 87-90 and 161-171 that this appeal is taken.

Claims 1-6, 8-11, 13-26, 28-30, 32-37, 87-90, 161, 162 and 164-171 stand rejected under 35 U.S.C. § 103 as being obvious over U.S. Patent No. 6,054,153 to Carr ("Carr") in view of U.S. Patent No. 4,522,835 to Woodruff ("Woodruff"), U.S. Patent No. 3,459,117 to Koch ("Koch") and U.S. Patent No. 6,042,859 to Shaklai ("Shaklai"). Claims 1, 2, 5-10, 12-15, 18-23, 25-29, 31-34, 36, 37, 87-90 and 161-171 stand rejected under 35 U.S.C. § 103 as being obvious over U.S. Patent No. 5,711,978 to Breen ("Breen") in view of Woodruff, Koch, Shaklai and DE 1935566 to Verbruggen ("Verbruggen").

4. STATUS OF AMENDMENTS

A Final Office Action was mailed on February 8, 2007. An Amendment and Response to the Office Action was previously filed on November 9, 2006. The Amendment and Response to the Office Action did amend claims 1, 22 and 161 by removing the phrase "the acts of" in the preamble. The Examiner entered these amendments in the Final Office Action of February 8, 2007.

5. SUMMARY OF CLAIMED SUBJECT MATTER

The present invention is directed to a method of manufacturing a modified atmosphere package (e.g., 10; FIG. 1). The independent claims comprise supplying a first package (e.g., 14; FIG. 1) including a non-barrier portion substantially permeable to oxygen (see, e.g., page 8, lines 20-22; page 10, lines 6-8). A retail cut of raw meat (e.g., 26; FIG. 1) is placed within the first package (e.g., 14; FIG. 1) with the meat (e.g., 26; FIG. 1) having meat pigment. The first package (e.g., 14; FIG. 1) is sealed (see, e.g., page 15, line 20). A second package (e.g., 12; FIG. 1) substantially impermeable to oxygen is supplied (see, e.g., page 9, lines 20-22). The first package (e.g., 14; FIG. 1) is covered with the second package (e.g., 12; FIG. 1) without sealing the second package (e.g., 12; FIG. 1) so as to create a pocket between the first and second packages (see, e.g., 14, 12; FIG. 1; page 9, line 31 – page 10, line 1). A mixture of gases is supplied into the pocket (see, e.g., page 10, line 28 – page 11, line 4).

Independent claim 1 further recites that the gas mixture comprises from about 0.1 to about 0.8 vol. % carbon monoxide (CO) and at least one other gas to form a low oxygen environment so as to form carboxymyoglobin on a surface of the raw meat (see, e.g., 26; FIG. 1; page 4; lines 19-22; page 17, lines 21-23). Independent claim 22 further recites that the gas mixture comprises from about 0.1 to about 0.8 vol. % CO and at least one other gas to form a low oxygen environment with the gas mixture being supplied so as to substantially convert the oxymyoglobin directly to carboxymyoglobin on a surface of the raw meat (see, e.g., 26; FIG. 1; page 4, line 31 – page 5, line 2; page 17, lines 21-23). Independent claim 161 further recites that the gas mixture comprises CO in an amount from about 0.3 vol. % to about 0.5 vol. % and at least one other gas to form a low oxygen environment so as to form carboxymyoglobin on a surface of the raw meat (see, e.g., 26; FIG. 1; page 11, lines 1-4).

Each of the independent claims includes removing oxygen from the pocket so as to sufficiently reduce an oxygen level therein so as to inhibit or prevent the formation of metmyoglobin on the surface of the raw meat (see, e.g., 26; FIG. 1; page 10, lines 17-30). The second package (e.g., 12; FIG. 1) is sealed wherein the carbon monoxide associated with the raw meat (e.g., 26; FIG. 1) within the first package (e.g., 14; FIG. 1) is adapted to be removable such that the color of the meat pigment is not fixed and turns brown in a natural time period upon removal of the second package (see, e.g., 12; FIG. 1; page 12, lines 2-11; page 21, lines 20-29).

6. GROUND S OF REJECTION TO BE REVIEWED ON APPEAL

I. Whether claims 1-6, 8-11, 13-26, 28-30, 32-37, 87-90, 161, 162 and 164-171 are obvious under 35 U.S.C. § 103 over U.S. Patent No. 6,054,153 to Carr in view of U.S. Patent No. 4,522,835 to Woodruff, U.S. Patent No. 3,459,117 to Koch and U.S. Patent No. 6,042,859 to Shaklai.

II. Whether claims 1, 2, 5-10, 12-15, 18-23, 25-29, 31-34, 36, 37, 87-90 and 161-171 are obvious under 35 U.S.C. § 103 over U.S. Patent No. 5,711,978 to Breen in view of Woodruff, Koch, Shaklai and Verbruggen.

7. GROUPING OF CLAIMS

Claims 1-37, 87-90 and 161-171 will stand or fall together.

8. ARGUMENT

The Appellants will discuss (1) the present invention, (2) the general case law of obviousness, (3) the reasons why a *prima facie* case has not been satisfied by either of the obviousness rejections and (4) additional evidence on why the pending claims are not obvious. To assist in explaining the present invention and showing the non-obviousness of the invention, the Appellants previously submitted evidence in the form of several 37 C.F.R. §1.132 declarations by (a) one of the co-inventors Mr. Gary R. DelDuca (Exhibits 1-6)¹; and (b) one skilled in the art of meat processing using modified atmosphere packaging -- Dr. Melvin C. Hunt (“the Hunt Declaration”)² (Exhibit 7).

I. Present Invention

The methods of manufacturing the modified atmosphere packages have several advantages: (a) the “seasoning” period of the raw meat may be reduced or eliminated; (b) the ability to obtain consistent blooming with cuts off pigment-sensitive meats (e.g., round bone) is improved; and (c) the ability to avoid “fixing” the color of the meat pigment to red. See, e.g., page 11, line 29 – page 12, line 15; page 13, lines 11-17 of the application; DelDuca Decl. ¶ 4 (Exhibit 1).

The “seasoning” period is the time period needed to diffuse the oxygen so that the meat has the ability to fully bloom. Page 3, lines 17-19 of the application; DelDuca Decl. ¶ 5. Trays, such as polystyrene foam trays, have a substantial amount of oxygen contained in its cellular structure that results in a time period of as long as about 5 to about 6 days to diffuse the oxygen contained in its cellular structure. Page 3, lines 21-23 of the application; DelDuca Decl. ¶ 5. If a foam tray is not used, the “seasoning” period can be reduced to one or two days. Page 3, lines 24-25 of the application; DelDuca Decl. ¶ 5. The reduction or elimination of the seasoning period “allows the meat to be displayed for retail sale much sooner than in existing low oxygen packaging systems.” Page 11, line 29 – page 12, line 2 of the application; DelDuca Decl. ¶ 5.

¹ The DelDuca declarations were respectfully submitted as exhibits in the Amendment and Response to Office Action Dated September 8, 2003; Amendment and Response to Office Action Dated February 17, 2004; Amendment and Response to Final Office Action Dated December 16, 2005; Response to Office Action Dated August 2, 2005; Amendment With RCE filed on May 25, 2006; and Amendment and Response to Office Action Dated August 10, 2006.

² The Hunt Declaration was submitted in this pending application as an exhibit in the Amendment and Response to Office Action Dated February 17, 2004.

Seasoning periods are not desired by the retailers or packers because of the “need to store and maintain the meat-filled packages for an extended duration before being opened for retail sale.” Page 3, lines 25-28 of the application; DelDuca Decl. ¶ 5.

One important aspect of the present invention is that the present invention does not “fix” the color of the meat pigment to red with its use of carbon monoxide (CO), but rather the meat pigment tends to turn brown in a natural time period after removal of the second package that is substantially impermeable to oxygen. See page 12, lines 2-12 of the application; DelDuca Third Decl. ¶ 3 (Exhibit 3). It is important to prevent the meat color from being “fixed” because it is unsafe (and potentially dangerous) to consume a piece of meat that has a bright red color that consumers associate with freshness, but is beyond the point of microbial soundness. See DelDuca Third Decl. ¶ 3. The term “fix” in this context does not mean that the color of meat pigment never changes to a brown color, but rather that the meat pigment does not turn brown in a natural time period after the meat pigment is exposed to atmosphere. *Id.*

The present invention “surprisingly allows the meat pigment to convert to metmyoglobin in a similar fashion as fresh, raw meat in a retail environment.” Page 12, lines 7-10 of the application; DelDuca Decl. ¶ 7 (Exhibit 1). Specifically, the color of the meat pigment after exposure to ambient temperature degrades in a fashion that is not beyond the point of microbial soundness, as if the CO had never been added to the modified packaging system. *Id.*

The meat used in the modified atmosphere packaging of the present invention substantially maintains its color during the shipping process because the package has a modified atmosphere in one embodiment that includes from about 0.1% to about 0.8 vol.% carbon monoxide. See DelDuca Decl. ¶ 8. In one method, after removal of the package that is substantially permeable to oxygen, the CO is lost to the atmosphere. See page 12, lines 2-6 of the application; DelDuca Decl. ¶ 8. The CO may be lost to the atmosphere through the package that includes a non-barrier portion that is substantially permeable to oxygen. *Id.* This allows the conversion of the carboxymyoglobin to oxymyoglobin by using the oxygen from the air. *Id.* The “gas mixture used in the modified atmosphere packages of the present invention, after removal, allows the carboxymyoglobin to convert to oxymyoglobin and then to metmyoglobin (brown) in a natural time period.” *Id.*

II. General Law on Obviousness

The Supreme Court in *KSR Int'l Co. v. Teleflex Inc.*, 550 U.S. ___, 2007 WL 1237837 (2007) (Exhibit 8) stated that the teaching, suggestion and motivation test is not to be rigidly applied, but did not apply a specific test to determine obviousness. Applying the *KSR Int'l* decision, the Federal Circuit in *Leapfrog Enterprises, Inc. v. Fisher-Price, Inc. and Mattel, Inc.* stated that “[a]n obviousness determination is not the result of a rigid formula disassociated from the consideration of the facts of a case.” No. 06-1402 (Fed. Cir. May 9, 2007) at page 7 (Exhibit 9). Indeed, the common sense of those skilled in the art demonstrates why some combinations would have been obvious where others would not. See *KSR Int'l Co. v. Teleflex Inc.* 550 U.S. ___, 2007 WL 1237837 at *12 (2007).

Prior to the *KSR Int'l Co.* decision, the teaching, suggestion and motivation test stated that all the limitations of a claim must be taught or suggested by the combined prior art references. M.P.E.P. § 2143.03 (citing *In re Royka*, 490 F.2d 981, 985, 180 U.S.P.Q. 580, 583 (C.C.P.A. 1974)). A *prima facie* case of obviousness requires three basic criteria:

First, there must be some suggestion or motivation, either in the references themselves or in the knowledge generally available to one of ordinary skill in the art, to modify the reference or to combine reference teachings. Second, there must be a reasonable expectation of success. Finally, the prior art reference (or references when combined) must teach or suggest all the claim limitations.

M.P.E.P. § 2143.

Obviousness cannot “be established using hindsight or in view of the teachings or suggestions of the invention.” *Ex parte Maguire*, No. 1999-1344, 2002 WL 1801466, at *4 (Bd. Pat. App. & Inter. 2002) (quoting *Para-Ordnance Mfg. Inc. v. SGS Importers Int'l Inc.*, 73 F.3d 1085, 1087, 37 U.S.P.Q.2d 1237, 1239 (Fed. Cir. 1995), *cert. denied*, 519 U.S. 822 (1996)). Further, the proposed modification cannot render the prior art “unsatisfactory for its intended purpose” nor can it “change the principle of operation” of a reference. M.P.E.P. § 2143.01 (citing *In re Gordon*, 733 F.2d at 900, 902, 221 U.S.P.Q. 1125, 1127 (Fed. Cir. 1984), and *In re Ratti*, 270 F.2d 810, 813, 123 U.S.P.Q. 349, 352 (C.C.P.A. 1959)).

The law of obviousness requires that a reference be considered as a whole, including those portions that teach away from the claimed invention. *Armament Sys. & Procedures v. Monadnock Lifetime Prods.*, No. 97-1174, 1998 U.S. App. LEXIS 20818, at *23-24 (Fed. Cir. 1998); see also M.P.E.P. § 2141.02 (stating that prior art must be considered in its entirety

including disclosures that teach away from the claims). Indicia of teaching away in a reference give insight into the question of obviousness. *Monarch Knitting Mach. Corp. v. Sulzer Morat GMBH*, 139 F.3d 877, 885, 45 U.S.P.Q.2d 1977, 1984 (Fed. Cir. 1998). A prior art reference may be considered to teach away when “a person of ordinary skill, upon reading the reference, would be discouraged from following the path set out in the reference, or would be led in a direction divergent from the path that was taken by the applicant.” *Id.* (quoting *In re Gurley*, 27 F.3d 551, 553, 31 U.S.P.Q.2d 1130, 1131 (Fed. Cir. 1994)).

The Examiner, of course, has the initial burden of establishing a *prima facie* basis to deny patentability to a claimed invention under any statutory provision. *In re Mayne*, 104 F.3d 1339, 1341, 41 U.S.P.Q.2d 1451, 1453 (Fed. Cir. 1997).

For at least the reasons stated below, Appellants respectfully submit that the Examiner has not set forth a *prima facie* case of obviousness under 35 U.S.C. § 103 and requests reversal of the Examiner’s 35 U.S.C. § 103 rejections.

III. A *Prima Facie* Case Has Not Been Presented With Respect To Independent Claims 1, 22 And 161

The pending independent claims (claims 1, 22 and 161) include, *inter alia*, (a) “a first package including a non-barrier portion substantially permeable to oxygen”, (b) “a second package substantially impermeable to oxygen”, (c) a low oxygen environment that includes from about 0.1 to about 0.8 vol.% CO or from about 0.3 to about 0.5 vol.% CO; and (d) “wherein the carbon monoxide associated with the raw meat within the first package is adapted to be removable such that the color of the meat pigment is not fixed and turns brown in a natural time period upon removal of the second package.” None of the applied references includes, *inter alia*, such limitations that are recited in independent claims 1, 22 and 161.

As acknowledged by the Examiner, U.S. Patent No. 6,054,153 to Carr and U.S. Patent No. 5,711,978 to Breen do not disclose, teach or suggest the use of CO. See pages 4 and 7 of the Office Action dated August 2, 2005. The Examiner applies a number of references – Woodruff, Koch, Shaklai and Verbruggen in an attempt to cure this deficiency in Carr and Breen. These other references do not disclose a packaging system having (a) “a first package including a non-barrier portion substantially permeable to oxygen”; and (b) “a second package substantially impermeable to oxygen” as recited in independent claims 1, 22 and 161.

It would not have been obvious to combine Carr or Breen in view of other references such as Koch, Woodruff, Shaklai and/or Verbruggen to arrive at the present invention. This erroneous conclusion by the Examiner ignores the understanding of those of ordinary skill in the art at the time of the present invention that CO “fixes” the color of the meat pigment and there would be no motivation to one of ordinary skill in the art for using CO in a modified atmosphere such as disclosed in Woodruff, Koch, Shaklai and/or Verbruggen with a meat-packaging system such as disclosed in Carr or Breen.

A. The Problems Of “Fixing” Color Are Known To Those Of Ordinary Skill In The Art

The problems of fixing meat color with CO, which can mask spoilage, are clearly known to those of ordinary skill in the art. See, e.g., Hunt Decl. ¶ 6 (Exhibit 7); DelDuca Decl. ¶ 6, 7 (Exhibit 1). The problem of fixing meat color with CO was described in a previously applied reference in this application to Sorheim et al.³ Furthermore, the United States Food and Drug Administration (FDA) has believed that the meat pigment color would be fixed using CO.⁴ Thus, the alleged “good” color (i.e., red color of fresh meat) disclosed in, for example, Woodruff is not a desirable attribute when the meat pigment remains such a color past its microbial soundness.

Additionally, Dr. Hunt, who has extensive experience in the processing of meat using modified atmosphere packaging, stated that “[t]he results of the testing [of the Pactiv’s improved ActiveTech® meat packaging system⁵] were surprising to me because it was understood by those skilled in the art that CO fixes (creates a stable form of myoglobin that could mask spoilage) the color of the meat pigment to red.” Hunt Decl. ¶ 6 (Exhibit 7). Pactiv’s improved ActiveTech® meat packaging system did not fix the color of the meat pigment as expected and Dr. Hunt stated

³ The applied reference was “The storage life of beef and pork packaged in an atmosphere with low carbon monoxide and high carbon dioxide” from *Meat Science* to Sorheim et al. (“Sorheim”)(Exhibit 10), which applied in the Office Action mailed on May 7, 2003. In particular, Sorheim disclosed that its meat packaging systems with a modified atmosphere of “0.4% CO/60% CO₂/40% N₂ had a bright stable red colour that lasted beyond the time of spoilage.” Abstract of Sorheim.

⁴ Exhibit 11 (In a 1962 letter, the FDA told a Whirlpool representative that it might need additional data “to establish that the treatment of meat would not serve to cause the meat to retain its fresh red color longer than meat not so treated” and that the FDA has a question “concerning possible deception of the consumer where treatment of the meat leads to longer retention of the fresh red color”, which was submitted in the Amendment and Response to Office Action Dated February 17, 2004); see also Hunt Decl. ¶ 6.

⁵ The process of Pactiv’s improved ActiveTech® meat-packaging system is one process that would be covered by the pending independent claims (claims 1, 22 and 161).

that “[t]his was a novel result and was not at all obvious due to the current and long standing thought that meat exposed to CO would develop a color that would mask spoilage.” See *id.*

Additionally, even individuals today believe that CO fixes the color of the meat pigment in meat-packaging applications. See, e.g., FDA Petition by Kalsec Foods (Exhibit 12) (see page 19 “the color-imparting effect of the carbon monoxide also masks the natural color change of meat due to aging and deceptively suggests freshness well past the microbial shelf life of the meat”; page 10 “[w]hen the oxygen in fresh meat packaging is displaced by carbon monoxide, the natural coloration provided by meat pigment is masked. Carbon monoxide binds firmly to myoglobin sites that otherwise would be bound gently by oxygen, forming carboxymyoglobin in place of oxymyoglobin.”); and an article entitled “FDA is urged to Ban Carbon-Monoxide-Treated Meat” (see page 1 ‘this meat [with CO] stays red and stays red and stays red’).

Thus, there is simply no motivation to combine Carr or Breen with Woodruff, Koch, Shaklai and/or Verbruggen in an attempt to address the problems solved by the present invention and to read on the pending claims.

B. The Applied References Of Shaklai, Koch, Woodruff And Verbruggen Do Not Teach Or Suggest That The Use Of CO Turns Meat Pigment Brown In A Natural Time Period

Based on the strong submitted evidence from the Appellants that those of ordinary skill in the art believed that CO “fixed” the color of the meat pigment at the time of the invention (i.e., that the meat pigment does not turn brown in a natural time period after the meat pigment is exposed to the atmosphere), the Examiner has attempted to apply a number of references allegedly stating otherwise. The Appellants will discuss these references and the reasons why they do not modify the belief before the Appellant’s invention that CO “fixed” the color of the meat pigment.

Specifically, the Examiner states that: (a) “Shaklai and Koch et al. provide evidence that the packages of either Carr [] or Breen [] will have CO [being] removably associated with the meat in a natural time period”; and (b) “the art of record does show that meat exposed to CO will brown within a natural time period after removal of CO and exposure to normal atmosphere.” Pages 5-6 of the Office Action dated August 10, 2006.

In addition to not disclosing, teaching or suggesting the claimed first and second packages, none of the references of Shaklai, Koch, Woodruff or Hermann, which are individually discussed in detail below, teaches or suggests the claimed limitation of “wherein the

carbon monoxide associated with the raw meat within the first package is adapted to be removable such that the color of the meat pigment is not fixed and turns brown in a natural time period upon removal of the second package.” Such limitations are specifically recited in independent claims 1, 22 and 161. Thus, there is no motivation to combine Shaklai, Koch, Woodruff and/or Verbruggen with Carr or Breen in the pending rejections.

i. Shaklai Teaches That CO “Fixes” The Color Of The Meat Pigment (I.e., Extends Color Life)

Since Shaklai teaches that CO “fixes” the color of the meat pigment after exposure to the atmosphere, there would be no motivation to one of ordinary skill in the art to combine Shaklai with (a) Carr, Koch and Woodruff; or (b) Breen, Koch, Woodruff and Verbruggen as in the pending rejections.

Specifically, Shaklai discloses exposing raw meat to an atmosphere consisting essentially of CO in which the meat is “completely immersed or saturated” with CO. See col. 5, lines 29-37. “More specifically, a cross-section of meat is completely immersed in or saturated to its core with carbon monoxide from the exposed surfaces through the entire cross-section (thickness) including its core region and retains the carbon monoxide until the meat is cooked. Thus, as stated above, the meat is preserved throughout its thickness.” Col. 5, lines 38-43 of Shaklai.

Shaklai continues by stating that “[p]ractically all of the carbon monoxide (over 99.9%) taken up by meat will be maintained as hemoglobin and myoglobin (Hb/Mb) bound forms.” Col. 5, lines 57-59. Shaklai also discloses that “[b]oth hemoglobin and myoglobin bind carbon monoxide much more strongly than oxygen.” Col. 5, lines 66-67. “It is thought that the mechanism for carbon monoxide preserving of meat is the much greater affinity of myoglobin for carbon monoxide than for oxygen.” Col. 6, lines 26-28 of Shaklai.

It is known to those of ordinary skill in the art that when hemoglobin in the red blood cells is exposed to CO, the CO has an affinity 200 times greater than oxygen does with hemoglobin.⁶ Therefore, one of ordinary skill in the art would expect that CO “fixes” the color of the meat pigment past its natural time period upon exposure to the atmosphere. DelDuca Third Decl. ¶ 4 (Exhibit 3). In other words, because of the hemoglobin’s high affinity towards

⁶ See, e.g., Color Atlas & Textbook of Hematology, Wm Platt, 2nd edition 1979 (Exhibit 13); DelDuca Third Decl. ¶ 4 (Exhibit 3).

CO, the pigment of the meat, prior to Appellants' invention, would not have been expected to degrade in a natural time period. *Id.*

The examples of Shaklai also support that the meat pigment is "fixed" beyond its natural time period. Specifically, Example 4 of Shaklai (mentioned at page 5 of the Office Action dated August 2, 2005) discloses that (a) meat treated with CO on day 14 had only a surface (less than 1 mm deep) being brown, while (b) meat treated with air was dark brown throughout. Col. 9, lines 40-50. Thus, it is clear that the meat pigment in Example 4 was "fixed" because it extended the color of meat pigment past its natural time period after being exposed to the atmosphere. This is further illustrated in Example 3 of Shaklai where the air-treated meat and CO-treated meat had different colors – the air-treated meat after 3 days was all brown and the CO-treated meat was a wine-red color. Col. 9, lines 10-19. Example 2 of Shaklai mentioned at page 5 of the Office Action dated August 2, 2005 also does not support that meat pigment is not "fixed" beyond its natural time period (air-treated samples were brown and CO-treated samples were a bright wine red after 24 hours). Col. 8, line 50-col. 9, line 5.

The Examiner asserts that Shaklai is being relied on as not fixing the color of the meat pigment surface. Pages 5, 7 and 8 of the Office Action dated August 10, 2006; Page 5 of the Office Action dated January 25, 2006. This ignores the evidence in the above examples that Shaklai discloses that the color of the meat pigment is fixed. There is no expectation in Shaklai that by applying the CO levels disclosed in Woodruff that the meat would brown in a natural time period, let alone the "reasonable expectation of success" asserted by the Examiner. See page 6 of the Office Action dated August 10, 2006.

Thus, because Shaklai discloses "fixing" the color of the meat pigment, there would be no motivation to one of ordinary skill in the art to combine Shaklai with (a) Carr, Koch and Woodruff or (b) Breen, Koch, Woodruff and Verbruggen as in the pending rejections because Shaklai discloses "fixing" the color of the meat pigment.

ii. Koch Does Not Teach Or Suggest That The Use Of CO Turns Meat Pigment Brown In A Natural Time Period

Since Koch does not teach or suggest that the use of CO turns meat pigment brown in a natural time period after removal of its CO-containing film, there would be no motivation to one of ordinary skill in the art to combine Koch with (a) Carr, Shaklai and Woodruff or (b) Breen, Shaklai, Woodruff and Verbruggen as in the pending rejections.

Specifically, Koch discloses wrapping meat with a CO-containing film such that CO is transferred from the film to contact the surface of the meat. See abstract. An object of Koch is to include a relatively small quantity of CO that is gradually released from the CO-containing film. Col. 2, lines 18-22. Koch discloses (a) covering primal cuts made at a slaughterhouse with a CO-containing film, (b) removing the CO-containing film at the retail outlet, and (c) cutting the primal cuts into individual steaks, roasts, etc. Col. 3, lines 4-8.

First, Koch does not disclose the exact weight of the primal cuts of meat. “Primal” cuts of meat at the time of the Koch disclosure (late 1960’s), however, generally refers to sections of meat from anywhere between about 50 and 150 or more lbs. DelDuca Third Decl. ¶ 6 (Exhibit 3). The term “subprimal” cuts of meat is used today and generally refers to cuts of meat from about 15 to about 20 lbs. *Id.* Thus, it is clear that the term primal cuts of meat in Koch refers to a large quantity of meat. *Id.*

Second, the disclosure of Shaklai with 100% CO (as compared to the small quantity of CO in Koch) took over 7 days to saturate a small piece of meat with CO. Specifically, in Example 3 of Shaklai, 0.5 to 1.5Kg (about 1.4 lbs to about 4.2 lbs) took 7 days upon exposure to 100% CO to turn the meat pigment to carboxymyoglobin. See col. 9, lines 11-28 of Shaklai and DelDuca Third Decl. ¶ 7. It would not be reasonable to one of ordinary skill in the art that a 50-150 lb piece of meat disclosed in Koch that had been exposed to a small quantity of CO would turn the non-surface meat pigments to carboxymyoglobin. DelDuca Third Decl. ¶ 7.

Therefore, when the primal cuts of meat of Koch were cut at the retail outlet into individual steaks and roasts, the meat pigments of such individual steaks and roasts had not been exposed to the CO from the CO-containing film. *Id.* It would be expected that the individually cut steaks and roasts sections of Koch that were not exposed to CO would degrade in a manner similar to other similar cuts of steaks and roasts that had also not been exposed to CO. DelDuca Third Decl. ¶ 8. Thus, Koch teaches that meat pigment in the form of individual steaks and roasts not exposed to CO in the CO-containing film would degrade in a similar manner of steaks and roasts not treated with CO. *Id.* Thus, Koch does not teach or suggest that the use of CO turns meat pigments brown in a natural time period after removal of the CO-containing film. *Id.*

In response to these arguments, the Examiner stated that Koch “is directed to the surface of the meat” and that Koch [] teach[es] that the surface treatment will result in the meat browning in a natural period.” Page 6 of the Office Action dated January 25, 2006. This ignores

the primal cuts to which Koch is directed, in which individual steaks and roast sections are not exposed to CO. Koch discloses “[w]hen the primal cuts arrive at the retail outlet, the covers are removed and the meat is cut into individual steaks, roasts, etc. which may be separately wrapped in conventional wrapping materials. It has been found that meat will release a saleable red color for as long as 10 days when covered with the cover herein described for the first seven days and with a conventional cover for the remaining days.”). Col. 3, lines 5-13 of Koch (underlining added); DelDuca Fifth Decl. ¶ 7.

In summary, Koch does not teach or suggest that the use of CO turns meat pigments brown in a natural time period after removal of the CO-containing film because it would not be reasonable that exposing a relatively small quantity of CO that is gradually released from the CO-containing film to a large quantity of meat (primal cuts) would expose CO to the non-surface meat pigments.

Since Koch does not teach or suggest that the use of CO turns meat pigment brown in a natural time period after removal of the CO-containing film, there would be no motivation to one of ordinary skill in the art to combine Koch with (a) Carr, Shaklai and Woodruff; or (b) Breen, Shaklai, Woodruff and Verbruggen as in the pending rejections.

iii. Woodruff Does Not Teach Or Suggest That The Use Of CO Turns Meat Pigment Brown In A Natural Time Period

The Examiner states that Woodruff “teach[es] that CO concentrations in the recited range of the applicant have been know to provide good color in meat during transportation and storage.” Page 5 of the Office Action dated August 10, 2006. The Examiner continues by stating that “Woodruff [] teaches surface contact of a meat with CO to maintain a red color”. *Id.*

Woodruff does not teach or suggest that the color of the meat pigment turns brown in a natural time period. DelDuca Fifth Decl. ¶ 4 (Exhibit 5). For example, Woodruff in Example 1 discloses a 0.5 lb. beefsteak that was exposed to 0.5% CO, which was nearly all absorbed two days later. See col. 4, lines 34-48; DelDuca Fifth Decl. ¶ 4. After being exposed in a modified atmosphere that included 16% oxygen, “the beefsteak retained its good red color, and the carboxymyoglobin color had penetrated no more deeply than it had at the end of the two days.” See col. 4, lines 49-54. This passage implies that the carboxymyoglobin color was still retained within the beefsteak after 6 days despite being exposed to an atmosphere with a generally similar amount of oxygen as in air (compare about 21% oxygen to 16% oxygen). DelDuca Fifth Decl. ¶

4. It would be expected to one skilled in the art that the beefsteak would turn brown in about 2-3 days, depending on the cut of meat. *Id.* Thus, this example clearly shows that the beefsteak of Woodruff in Example 1 did not turn brown in a natural time period, but rather “fixed” the color of the meat pigment. *Id.* Similarly, in Example 1 of Woodruff, a 0.5 lb. beefsteak exposed to 2.5% CO under similar conditions also retained its good color after 6 days. See col. 4, line 55-col. 5, line 6; DelDuca Fifth Decl. ¶ 4.

None of the other examples of Woodruff supports a modified atmosphere package wherein the CO associated with the raw meat is adapted to be removed such that the color of the meat pigment is not fixed and turns brown in a natural time period. DelDuca Fifth Decl. at ¶ 5. Rather, the other examples of Woodruff generally disclose the condition of the meat pigment while being stored in a modified atmosphere containing CO. *Id.* In summary, Woodruff does not disclose, teach or suggest that the use of CO on meat pigment turns brown in a natural time period, but rather Woodruff teaches and suggests “fixing” the color of the meat pigment in Example 1. *Id.* at 6.

Since Woodruff does not teach or suggest that the use of CO turns meat pigment brown in a natural time period, there would be no motivation to one of ordinary skill in the art to combine Woodruff with (a) Carr, Shaklai and Koch or (b) Breen, Shaklai, Koch and Verbruggen as in the pending rejections.

iv. Verbruggen Does Not Teach Or Suggest That The Use Of CO Turns Meat Pigment Brown In A Natural Time Period

The other applied reference (Verbruggen) has the same deficiencies of Woodruff. The Examiner briefly discusses Verbruggen as follows: “Verbruggen provides further evidence of the conventionality of utilizing a carbon dioxide and carbon monoxide mixture for preserving meat.” Page 5 of the Office Action dated August 10, 2006. Verbruggen, thus, does not teach or suggest that the use of CO turns meat pigment brown in a natural time period and there would be no motivation to one of ordinary skill in the art to combine Verbruggen with (a) Carr, Shaklai, Koch and Woodruff; or (b) Breen, Shaklai, Koch and Woodruff as in the pending rejections.

Thus, the Appellants believe that a *prima facie* case has not been presented with Carr, Breen, Woodruff, Koch, Shaklai, Verbruggen or any combination thereof.

IV. Evidence of Non-Obviousness of Independent Claims 1, 22 And 161

Assuming, *arguendo*, that a *prima facie* case has been presented (which Appellants strongly believe is not the case), the Appellants previously submitted evidence of non-obviousness including the DelDuca Declarations (Exhibits 1-6) and the Hunt Declaration (Exhibit 7). Secondary consideration such as commercial success, long felt but unsolved needs, failure of others, etc. may be used to give light to the circumstances surrounding the origin of the subject and, thus, may be used to rebut a *prima facie* case of obviousness. See *Graham v. John Deere Co. of Kansas City*, 383 U.S. 1, 17-18 (1966).

A. CO Not Allowed With Fresh Meat In The United States Since At Least 1962

Carbon monoxide (CO) has not been allowed to be used with fresh meat in the United States for about 40 years.^{7,8} The Food and Drug Administration (“FDA”) regulation that currently prevents using CO with meat-packaging systems in the United States is 21 C.F.R. § 173.350.

The food additive combustion product gas may be safely used in the processing and packaging of the foods designated in paragraph (c) of this section for the purpose of removing and displacing oxygen...(b) The food additive meets the following specifications: (1) Carbon monoxide content not to exceed 4.5 percent by volume...(c) It [carbon monoxide] is used or intended for use to displace or remove oxygen in the processing, storage, or packaging of beverage products and other food, except fresh meats.

Exhibit 16 (emphasis added); see also DelDuca Decl. ¶ 9.

The concern of the FDA is believed to be that CO fixes the fresh meat color to a degree that allows the retailer to sell meat that looks good (a bright red color), but is unsafe and potentially dangerous to consume because it has unacceptable levels of bacteria. DelDuca Decl.

⁷ 21 U.S.C. § 121.1060 was first promulgated on August 2, 1961 (Exhibit 14) and permitted the use of combustion product gas containing up to 4.5% CO for use “to displace or remove oxygen or remove oxygen in the processing, storage, or packaging of citrus products, vegetable fats and vegetable oils, coffee, and wine.” In December 14, 1962, 21 U.S.C. § 121.1060 (Exhibit 15) was amended to exclude fresh meats. In March of 1977, 21 U.S.C. § 121.1060 was re-designated as 21 C.F.R. § 173.350. Both Exhibits 14 and 15 were submitted in the Amendment and Response to Office Action Dated May 7, 2003.

⁸ See also DelDuca Decl. ¶ 9.

¶ 10 (Exhibit 1).⁹ This act of fixing the meat color to a bright red color is referred to as “economic adulteration.” *Id.*

B. CO Now Allowed In Pactiv’s Improved ActiveTech® Meat-Packaging System

After about 40 years of not allowing CO to be used with fresh meats in the United States, the Appellants came up with novel approaches of using CO in modified atmosphere packaging (MAP) systems that avoided the concerns of “fixing” the meat color. DelDuca Decl. ¶ 11 (Exhibit 1). The assignee of the patent application (Pactiv Corporation) gave notice to the FDA in August of 2001 of a specific embodiment and process, and evidence supporting Pactiv’s conclusion that CO as used is GRAS (generally recognized as safe). See Exhibit B of Hunt Decl. (Exhibit 7); See DelDuca Decl. ¶ 12. The MAP system in Pactiv’s GRAS notice may be used for packaging meats such as fresh cuts of case-ready muscle meat and ground case-ready meat to maintain wholesomeness, provide flexibility in distribution, and prevent losses due to spoilage at retail sale. See page 5 of Exhibit B of Hunt Decl.; DelDuca Decl. ¶ 12.

The specific MAP system that was presented in the GRAS notice used 0.4 vol. % CO in a meat-packaging system and was referred to as Pactiv’s improved ActiveTech® meat-packaging system. See page 5 of Exhibit B of Hunt Decl. (Exhibit 7); DelDuca Decl. ¶ 13. Unlike Pactiv’s traditional ActiveTech® meat-packaging system, Pactiv’s improved ActiveTech® meat-packaging system used CO. Pactiv’s improved ActiveTech® meat-packaging system includes meats being placed in polystyrene trays and covered with oxygen-permeable, polyvinyl chloride (“PVC”) overwraps. Page 6 of Exhibit B of Hunt Decl.; DelDuca Decl. ¶ 13. The wrapped trays of meat are then placed in an outer barrier bag. Ambient air is removed and replaced with a blend of 0.4% CO, 30% carbon dioxide, and the balance being nitrogen. Page 8 of Exhibit B of Hunt Decl.; DelDuca Decl. ¶ 13. The myoglobin of the meat converts from oxymyoglobin to carboxymyoglobin (red). Page 9 of Exhibit B of Hunt Decl.; DelDuca Decl. ¶ 13. The meat maintains its red color while in storage until the package is opened for retail display by removing the outer barrier bag. *Id.*

⁹ See, e.g., Exhibit 11 (In a 1962 letter, the FDA told a Whirlpool representative that it might need additional data “to establish that the treatment of meat would not serve to cause the meat to retain its fresh red color longer than meat not so treated” and that the FDA has a question “concerning possible deception of the consumer where treatment of the meat leads to longer retention of the fresh red color.”)

The package will lose CO to the atmosphere and, thus, at retail display will not have CO. DelDuca Decl. ¶ 13. Once in retail display, the meat's myoglobin begins its natural conversion to metmyoglobin (brown). Page 9 of Exhibit B of Hunt Decl.; DelDuca Decl. ¶ 13. The CO used in the MAP system did not mask the spoilage or extend the color life beyond the point of wholesomeness (i.e., the point of microbial soundness). Pages 9, 10 of Exhibit B of Hunt Decl. DelDuca Decl. ¶ 13.

The FDA stated that it had no questions regarding Pactiv's conclusion about Pactiv's improved ActiveTech® meat-packaging system using 0.4 vol.% CO being GRAS because of the evidence presented by Pactiv in its notice. Exhibit 17 at page 1; DelDuca Decl. ¶ 14. Specifically, the FDA stated the following: "Based on the data and information reviewed, Pactiv's GRAS panel conclude[d] that CO, when produced in accordance with current good manufacturing practice and meeting appropriate food grade specifications, is GRAS, through scientific procedures under the conditions of its intended use." Exhibit 17 at page 1. This FDA review allows Pactiv to use CO with fresh meat in its application. DelDuca Decl. ¶ 14. It is believed to be the first system to overcome the prohibition of CO with fresh meat in the United States in the last 40 years. *Id.*

Thus, a problem of fixing meat color with CO that was recognized for at least the last 40 years was overcome by Pactiv's improved meat-packaging system and process of the same. See, e.g., Exhibits 11, 17; Exhibit B of the Hunt Decl. The Pactiv process in the GRAS notice is an example of one process that would be covered by claims 1, 22 and 161 of the pending application. Specifically, the Pactiv process included (a) a first package that included a PVC overwrap, (b) a retail cut of raw meat within the first package that was sealed, (c) a second outer barrier bag that covered the first package, (d) a gas mixture including 0.4 vol. % CO that formed a low oxygen environment resulting in carboxymyoglobin on a surface of the raw meat, and (e) sealing the second package.

The Examiner has asserted for the first time that Pactiv's improved ActiveTech® meat packaging system using 0.4 vol.% CO is not commensurate in scope with independent claims 1 and 22.¹⁰ See Office Action dated February 8, 2007. The Appellants disagree. First, the claimed amount of 0.1 to about 0.8 vol. % CO is clearly within the scope of the Pactiv's

¹⁰ The Examiner apparently did not have any problems with the scope of independent claim 161 (using from about 0.3 to about 0.5 vol. % CO).

improved ActiveTech® meat packaging system using 0.4 vol.% CO. Additionally, there are dependent claims that further narrow the amount of CO being used (see, e.g., claim 21 with 0.1 to about 0.5 vol.% CO being claimed).

C. The Pactiv Improved ActiveTech® Meat-Packaging System and Process Using CO Address a Long-Felt Need

The Federal Circuit has stated that if an invention unexpectedly solved longstanding problems, it supports the conclusion of nonobviousness. See, e.g., *Hybritech Inc. v. Monoclonal Antibodies, Inc.*, 802 F.2d 1367, 1382 (Fed. Cir 1986); *WMS Gaming Inc. v. Int'l Game Tech.*, 184 F.3d 1339, 1359 (Fed. Cir. 1999).

The process of manufacturing Pactiv's improved ActiveTech® meat-packaging system, which is an example of a process that would be covered under independent claims 1, 22 and 161 of the pending application, addressed such a long-felt need in the meat-packaging industry. "Prior to Pactiv's [improved] ActiveTech® meat packaging system using 0.4 vol.% CO, there was a need in the industry to provide a solution that: (a) reduced the seasoning period (the critical time meat is exposed to low partial pressures of oxygen, which can seriously damage the pigment chemistry); (b) formed consistently a normal bloomed color with meats whose pigment is sensitive to metmyoglobin formation; and (c) avoided the fixing of too stable of a meat color, which can be unsafe and potentially dangerous, if the color stability was greater than the shelf life (microbial soundness) of the product." Hunt Decl. ¶ 7 (Exhibit 7). "Such a solution was especially desirable for a centralized packaging facility where the meat would be shipped to distant locations." *Id.* "Pactiv's [improved] ActiveTech® meat packaging system using 0.4 vol. % CO was a new and novel approach that addressed these technological needs." *Id.* Dr. Hunt stated that the results of the testing of Pactiv's ActiveTech® meat-packaging system were surprising. See *id.* at ¶¶ 5, 6.

Thus, since Pactiv's improved ActiveTech® meat-packaging process surprisingly addressed a long-felt need, this is further evidence that the independent claims of the present application are not obvious over the applied references.

In response to this evidence on long-felt need, the Examiner asserted that "[i]t is notoriously well known in the art [from the applied references] that a red colored meat at the retail outlet is most desired. It was also known that meat exposed to CO in a modified atmosphere environment would provide the meat with a red color after the meat was removed

from the modified atmosphere environment.” Page 10 of the Office Action dated August 10, 2006. This clearly ignores the understanding of those skilled in the art prior to Appellant’s invention that CO “fixed” the color of the meat pigment, which is discussed in detail above.

D. The Pactiv Improved ActiveTech® Meat Packaging System and the Process of Using the Same is Commercially Successful

The Federal Circuit has also stated that “[c]ommercial success is . . . a strong factor favoring non-obviousness.” See, e.g., *Akzo N.V. v. U.S. Int’l Trade Comm’n*, 808 F.2d 1471, 1481 (Fed. Cir. 1986); see also *Gambro Lundia AB v. Baxter Healthcare Corp.*, 110 F.3d 1573, 1579-80 (Fed. Cir. 1997).

Pactiv and its predecessor Tenneco Packaging Inc. (herein “Pactiv”) have sold modified atmosphere packaging systems beginning in 1998 (the traditional ActiveTech® meat packaging system). DelDuca Sixth Decl. ¶ 4 (Exhibit 6). The Pactiv traditional ActiveTech® meat packaging system includes meats being placed in polystyrene trays and covered with oxygen-permeable, PVC overwraps. *Id.* The wrapped trays of meat are then placed in an outer barrier bag. Ambient air is removed and replaced with a blend of 30 vol. % carbon dioxide, and the balance being nitrogen. *Id.*

Beginning in March of 2002, Pactiv began offering for sale an improved ActiveTech® meat-packaging system. DelDuca Sixth Decl. ¶ 5. Pactiv’s improved ActiveTech® meat-packaging system includes meats being placed in polystyrene trays and covered with oxygen-permeable, PVC overwraps. *Id.* The wrapped trays of meat are then placed in an outer barrier bag. *Id.* Ambient air is removed and replaced with a blend of 0.4 vol. % carbon monoxide (CO), 30 vol. % carbon dioxide, and the balance being nitrogen. *Id.*

The modified atmosphere used in Pactiv’s improved ActiveTech® meat-packaging system differs from the modified atmosphere used in the Pactiv’s traditional ActiveTech® meat-packaging system. DelDuca Sixth Decl. ¶ 6. Specifically, Pactiv’s improved ActiveTech® meat packaging system uses 0.4 vol.% CO, while Pactiv’s traditional ActiveTech® meat-packaging system does not use CO. *Id.* Because of the addition of CO, the equipment used in Pactiv’s improved ActiveTech® meat-packaging system may vary slightly as compared to Pactiv’s traditional ActiveTech® meat-packaging system. *Id.* Specifically, a mixer may be added to Pactiv’s improved ActiveTech® meat-packaging system to mix the CO, carbon dioxide, and

nitrogen. *Id.* Additionally, a CO gas recovery hood and safety features may also be included in Pactiv's improved ActiveTech® meat-packaging system.

The purchasers of either Pactiv's improved ActiveTech® meat-packaging system or Pactiv's traditional ActiveTech® meat-packaging system receive a license for the process and the knowledge to run such a process. DelDuca Sixth Decl. ¶ 7 (Exhibit 4). Pactiv allows its customers to use its oxygen-absorber dispensing-machine at no cost. *Id.* The remaining machinery used to perform either Pactiv's improved ActiveTech® meat packaging system or Pactiv's traditional ActiveTech® meat packaging system is purchased by the customer. *Id.* Typically, this remaining machinery is sold by Pactiv to its customers. *Id.* The customers also typically purchase the oxygen absorbers, trays, and film from Pactiv. *Id.*

Sales of Pactiv's traditional ActiveTech® meat-packaging system were decreasing in 2000 and 2001. DelDuca Sixth Decl. ¶ 8. The sales of Pactiv's improved ActiveTech® meat packaging system, however, have substantially increased since its introduction in March of 2002. *Id.* The sales of Pactiv's improved ActiveTech® meat-packaging system have been commercially successful with sales numbers of about or over 6 million dollars in each of the years since 2003. *Id.* These sales include the total of the purchased licenses, the purchased remaining machinery, and supplies (which include oxygen absorbers, activator fluid, and film). *Id.*

Since March of 2002, both Pactiv's improved ActiveTech® meat-packaging system and Pactiv's traditional ActiveTech® meat-packaging system have been available for sale. DelDuca Sixth Decl. ¶ 9. Since March 2002, no customer has purchased Pactiv's traditional ActiveTech® meat-packaging system. *Id.* In fact, every customer still practicing Pactiv's technology has converted its traditional ActiveTech® meat-packaging system into Pactiv's improved ActiveTech® meat-packaging system. *Id.* Thus, to my knowledge no customer is still practicing Pactiv's traditional ActiveTech® meat-packaging system. *Id.* It can be concluded that these customers prefer the Pactiv's improved ActiveTech® meat packaging system over Pactiv's traditional ActiveTech® meat-packaging system. *Id.* The cost of Pactiv's improved ActiveTech® meat-packaging system versus Pactiv's traditional ActiveTech® meat-packaging system is fractionally more expensive. *Id.* Thus, the commercial success of Pactiv's improved ActiveTech® meat-packaging system cannot be attributed to a cost advantage. *Id.*

Since 2002, there has been no increase in the number of sales personnel from Pactiv who are responsible for sales of Pactiv's improved ActiveTech® meat packaging system. DelDuca Sixth Decl. ¶ 10. In fact, the number of sales personnel who are responsible for sales of the Pactiv's improved ActiveTech® meat-packaging system have decreased since 2002. *Id.* There has been little or no advertising directed to sales of Pactiv's improved ActiveTech® meat packaging system since 2002. *Id.* The amount of advertising, if any, has not increased since 2002 and likely has decreased substantially from that directed to Pactiv's traditional ActiveTech® meat-packaging system. *Id.* Thus, the commercial success of Pactiv's improved ActiveTech® meat-packaging system cannot be attributed to increased marketing/advertising. *Id.*

The process of manufacturing using Pactiv's improved ActiveTech® meat-packaging system is an example of a process that would be covered under independent claims 1, 22 and 161 of the present application. DelDuca Sixth Decl. ¶ 11.

One of the arguments raised by the Examiner with the submitted evidence in commercial success was that "it is not clear if the claimed invention resulted in the commercial success or whether other factors contributed to the success, such as increase[d] advertising/marketing." Page 10 of the Office Action dated August 10, 2006. It is clear from the submitted evidence that Pactiv's improved ActiveTech® meat packaging is commercially successful and that factors such as increased advertising/marketing were not the cause of its success.

In the latest response (Final Office Action dated February 8, 2007), the Examiner also stated that "sales of improved package could have been a result of attractive pricing on part of the Pactiv [C]orporation, where the company could have offered the newer system of packaging for relatively very small increase in price which would still be fractionally higher than the original (Declaration page 4) over the traditional package." This statement is nonsensical. In the DelDuca Sixth Declaration (Exhibit 6), the Appellants submitted evidence that the Pactiv's improved ActiveTech® meat packaging (which would be covered by the existing claims) was "fractionally more expensive" than Pactiv's traditional ActiveTech® meat-packaging system. Thus, cost was not an issue for a customer selecting Pactiv's improved ActiveTech® meat-packaging system over Pactiv's traditional ActiveTech® meat-packaging system.

The Examiner also stated that the "company might have provided free training and license to use the product in order to boost the sales" and "[i]t has been noted that Pactiv allows

its customers to use oxygen absorber machines at no cost . . . , however it is not clear when this practice of no charge use of oxygen absorber was implemented, as it could also be a factor in altering sales.” Pages 4 and 5 of the Final Office Action dated February 8, 2007. The Appellants disagree and provided the following evidence in the DelDuca Sixth Declaration at paragraph 7:

The purchasers of either Pactiv’s improved ActiveTech® meat packaging system or Pactiv’s traditional ActiveTech® meat packaging system receive a license for the process and the knowledge to run such a process. Pactiv allows its customers to use its oxygen-absorber dispensing-machine at no cost. The remaining machinery used to perform either Pactiv’s improved ActiveTech® meat packaging system or Pactiv’s traditional ActiveTech® meat packaging system is purchased by the customer. Typically, this remaining machinery is sold by Pactiv to its customers. The customers also typically purchase the oxygen absorbers, trays, and film from Pactiv.

As discussed above, there was no cost to Pactiv’s customers on its oxygen-absorber dispensing-machine and Pactiv provided free training and the license to use the product for both Pactiv’s improved ActiveTech® meat packaging system and Pactiv’s traditional ActiveTech® meat packaging system. Thus, these were not factors in the customers choosing Pactiv’s improved ActiveTech® meat packaging system over Pactiv’s traditional ActiveTech® meat packaging system.

Therefore, in addition to the applied references not presenting a *prima facie* case, the Appellants also believe that the pending claims are allowable because of the compelling evidence of non-obviousness. Therefore, independent claims 1, 22 and 161 are not obvious in view of Carr, Breen, Woodruff, Koch, Shaklai and Verbruggen or any combination thereof and, thus, should be in a condition for allowance and the Appellants request reversal of the Examiner’s 35 U.S.C. § 103 rejections.

9. CONCLUSION

For the reasons set forth above, Appellants respectfully submit that the Examiner’s rejections fail to present a *prima facie* case of obviousness under 35 U.S.C. § 103. Additionally, even if a *prima facie* case has been presented (which Appellants strongly believe is not the case), the overwhelming evidence of non-obviousness rebuts any *prima facie* case of obviousness. Based upon the arguments submitted above, Appellants respectfully solicit the reversal of the

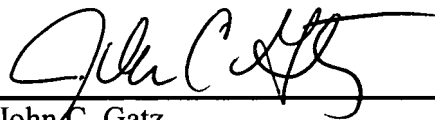
Examiner's 35 U.S.C. § 103 rejections of claims 1-37, 87-90 and 161-171 on at least the grounds noted above.

The Appellants note that the fee of \$500.00 required by 37 C.F.R. §41.20(b)(2) has already been paid. The Appellants request that the fee of \$500.00 for the Notice of Appeal filed under 37 C.F.R. §41.20(b)(1) be deducted from Nixon Peabody LLP Deposit Account No. 50-4181.

The Commissioner is also hereby authorized to charge deposit account No. 50-4181 (Attorney Docket No. 247097-001080USPT) for any additional fees inadvertently omitted which may be necessary now or during the pendency of this application, except for the issue fee.

May 18, 2007
Date

Respectfully submitted,

A handwritten signature in black ink, appearing to read "John C. Gatz", is written over a horizontal line.

John C. Gatz
Registration No. 41,774
NIXON PEABODY LLP
161 North Clark Street
Suite 4800
Chicago, Illinois 60601
(312) 425-3900 - Telephone

Attorney for Appellants

10. APPENDIX OF CLAIMS ON APPEAL

Listing of Claims:

1. A method of manufacturing a modified atmosphere package, the method comprising:

supplying a first package including a non-barrier portion substantially permeable to oxygen;

placing a retail cut of raw meat within the first package, the meat having meat pigment;

sealing the first package;

supplying a second package substantially impermeable to oxygen;

covering the first package with the second package without sealing the second package so as to create a pocket between the first and second packages;

supplying a mixture of gases into the pocket, the gas mixture comprising from about 0.1 to about 0.8 vol.% carbon monoxide and at least one other gas to form a low oxygen environment so as to form carboxymyoglobin on a surface of the raw meat;

removing oxygen from the pocket so as to sufficiently reduce an oxygen level therein so as to inhibit or prevent the formation of metmyoglobin on the surface of the raw meat; and

sealing the second package, wherein the carbon monoxide associated with the raw meat within the first package is adapted to be removable such that the color of the meat pigment is not fixed and turns brown in a natural time period upon removal of the second package.

2. The method of claim 1 further including supplying an oxygen scavenger.

3. The method of claim 1 further including supplying an oxygen scavenger, activating the oxygen scavenger with an oxygen scavenger accelerator, and positioning the oxygen scavenger external to the first package such that the oxygen scavenger is capable of absorbing oxygen within the pocket, the activated oxygen scavenger aggressively absorbing any residual oxygen in the modified atmosphere package.

4. The method of claim 3, wherein the activated oxygen scavenger reduces the oxygen level within the modified atmosphere package to approximately zero percent in less than

about 24 hours.

5. The method of claim 1, wherein the oxygen level of the pocket is less than 1,000 ppm.

6. The method of claim 5, wherein the oxygen level of the pocket is less than about 500 ppm.

7. The method of claim 1, wherein removing oxygen from the pocket includes evacuating the pocket.

8. The method of claim 1, wherein removing oxygen from the pocket includes flushing the pocket with the gas mixture.

9. The method of claim 1, wherein the gas mixture further comprises nitrogen, carbon dioxide or the combination thereof.

10. The method of claim 1, wherein the gas mixture further consists essentially of nitrogen, carbon dioxide or the combination thereof.

11. The method of claim 1, wherein the gas mixture consists essentially of from about 0.1 to about 0.8 vol. % carbon monoxide, from about 40 to about 80 vol.% nitrogen and from about 20 to about 60 vol.% carbon dioxide.

12. The method of claim 1, wherein the gas mixture consists of from about 0.1 to about 0.8 vol.% carbon monoxide with the remainder carbon dioxide.

13. The method of claim 1 further including removing the second package from the first package before retailing.

14. The method of claim 1 further including removing the second package from the first package so as to allow the raw meat to be exposed to ambient atmosphere, the raw meat having color degradation similar to a fresh cut of the same raw meat.

15. The method of claim 1, wherein the second package is adapted to be removable from at least a portion of the first package without destroying the first package.

16. The method of claim 1 further including placing the retail cut of raw meat on a foam tray.

17. The method of claim 1, wherein the non-barrier portion comprises a polyolefin or a polyvinyl chloride overwrap.

18. The method of claim 1, wherein the gas mixture is supplied to the pocket such that the oxymyoglobin substantially converts directly to carboxymyoglobin.

19. The method of claim 1, wherein the oxymyoglobin substantially converts to deoxymyoglobin before the gas mixture is supplied to the pocket so as to convert deoxymyoglobin directly to carboxymyoglobin.

20. The method of claim 1, wherein the gas mixture comprises from about 0.3 to about 0.5 vol.% carbon monoxide.

21. The method of claim 1, wherein the gas mixture comprises from about 0.1 to about 0.5 vol.% carbon monoxide.

22. A method of manufacturing a modified atmosphere package, the method comprising:

supplying a first package including a non-barrier portion substantially permeable to oxygen;

placing a retail cut of raw meat within the first package, the meat having meat pigment;

sealing the first package;

supplying a second package substantially impermeable to oxygen;

covering the first package with the second package without sealing the second package so as to create a pocket between the first and second packages;

supplying a mixture of gases into the pocket, the gas mixture comprising from about 0.1 to about 0.8 vol.% carbon monoxide and at least one other gas to form a low oxygen environment, the gas mixture being supplied so as to substantially convert the oxymyoglobin directly to carboxymyoglobin on a surface of the raw meat;

removing oxygen from the pocket so as to reduce an oxygen level sufficiently therein so as to inhibit or prevent the formation of metmyoglobin on the surface of the raw meat; and

sealing the second package, wherein the carbon monoxide associated with the raw meat within the first package is adapted to be removable such that the color of the meat pigment is not fixed and turns brown in a natural time period upon removal of the second package.

23. The method of claim 22 further including supplying an oxygen scavenger.

24. The method of claim 22 further including supplying an oxygen scavenger, activating the oxygen scavenger with an oxygen scavenger accelerator, and positioning the oxygen scavenger external to the first package such that the oxygen scavenger is capable of

absorbing oxygen within the pocket, the activated oxygen scavenger aggressively absorbing any residual oxygen in the modified atmosphere package.

25. The method of claim 22, wherein the oxygen level of the pocket is less than 1,000 ppm.

26. The method of claim 25, wherein the oxygen level of the pocket is less than about 500 ppm.

27. The method of claim 22, wherein removing oxygen from the pocket includes evacuating the pocket.

28. The method of claim 22, wherein removing oxygen from the pocket includes flushing the pocket with the gas mixture.

29. The method of claim 22, wherein the gas mixture further comprises nitrogen, carbon dioxide or the combination thereof.

30. The method of claim 22, wherein the gas mixture consists essentially of from about 0.1 to about 0.8 vol.% carbon monoxide, from about 40 to about 80 vol.% nitrogen and from about 20 to about 60 vol.% carbon dioxide.

31. The method of claim 22, wherein the gas mixture consists of from about 0.1 to about 0.8 vol.% carbon monoxide with the remainder carbon dioxide.

32. The method of claim 22 further including removing the second package from the first package before retailing.

33. The method of claim 22 further including removing the second package from the first package so as to allow the raw meat to be exposed to ambient atmosphere, the raw meat having color degradation similar to a fresh cut of the same raw meat.

34. The method of claim 22, wherein the second package is adapted to be removable from at least a portion of the first package without destroying the first package.

35. The method of claim 22 further including placing the retail cut of raw meat on a foam tray and the non-barrier portion comprises a polyolefin or a polyvinyl chloride overwrap.

36. The method of claim 22, wherein the gas mixture comprises from about 0.3 to about 0.5 vol.% carbon monoxide.

37. The method of claim 22, wherein the gas mixture comprises from about 0.1 to about 0.5 vol.% carbon monoxide.

38-86. Cancelled.

87. The method of claim 1, wherein after sealing the first and second packages, the modified atmosphere package is modified so as to allow the raw meat to be exposed to ambient atmosphere.

88. The method of claim 87, wherein the modified atmosphere package is modified by having the second package removed from at least a portion of the first package so as to allow the raw meat to be exposed to ambient atmosphere.

89. The method of claim 22, wherein after sealing the first and second packages, the modified atmosphere package is modified so as to allow the raw meat to be exposed to ambient atmosphere.

90. The method of claim 89, wherein after sealing the first and second packages, the modified atmosphere package is modified so as to allow the raw meat to be exposed to ambient atmosphere.

91-160. Cancelled.

161. A method of manufacturing a modified atmosphere package, the method comprising:

supplying a first package including a non-barrier portion substantially permeable to oxygen;

placing a retail cut of raw meat within the first package, the meat having meat pigment;

wrapping the first package with a polyolefin or a polyvinyl chloride overwrap;

supplying a second package substantially impermeable to oxygen;

covering the first package with the second package without sealing the second package so as to create a pocket between the first and second packages;

supplying a mixture of gases into the pocket, the gas mixture comprising from about 0.3 to about 0.5 vol.% carbon monoxide and at least one other gas to form a low oxygen environment so as to form carboxymyoglobin on a surface of the raw meat; and

removing oxygen from the pocket so as to sufficiently reduce an oxygen level therein so as to inhibit or prevent the formation of metmyoglobin on the surface of the raw meat.

sealing the second package wherein the carbon monoxide associated with the raw meat

within the first package is adapted to be removable such that the color of the meat pigment is not fixed and turns brown in a natural time period upon removal of the second package.

162. The method of claim 161 further including supplying an oxygen scavenger.

163. The method of claim 161, wherein removing oxygen from the pocket includes evacuating the pocket.

164. The method of claim 161, wherein removing oxygen from the pocket includes flushing the pocket with the gas mixture.

165. The method of claim 161, wherein the gas mixture further comprises nitrogen, carbon dioxide or the combination thereof.

166. The method of claim 161, wherein the gas mixture further consists essentially of nitrogen, carbon dioxide or the combination thereof.

167. The method of claim 161 further including removing the second package from the first package before retailing.

168. The method of claim 161 further including removing the second package from the first package so as to allow the raw meat to be exposed to ambient atmosphere, the raw meat having color degradation similar to a fresh cut of the same raw meat.

169. The method of claim 161, wherein the second package is adapted to be removable from at least a portion of the first package without destroying the first package.

170. The method of claim 161, wherein after wrapping the first package and sealing the second package, the modified atmosphere package is modified so as to allow the raw meat to be exposed to ambient atmosphere.

171. The method of claim 170, wherein the modified atmosphere package is modified by having the second package removed from at least a portion of the first package so as to allow the raw meat to be exposed to ambient atmosphere.

172-189. Cancelled.

11. EVIDENCE APPENDIX

- Exhibit 1 – Declaration of Mr. Gary DelDuca
- Exhibit 2 – Second Declaration of Mr. Gary DelDuca
- Exhibit 3 – Third Declaration of Mr. Gary DelDuca
- Exhibit 4 – Fourth Declaration of Mr. Gary DelDuca
- Exhibit 5 – Fifth Declaration of Mr. Gary DelDuca
- Exhibit 6 – Sixth Declaration of Mr. Gary DelDuca
- Exhibit 7 – Declaration of Dr. Melvin C. Hunt
 - Exhibit A – Curriculum Vitae of Dr. Melvin C. Hunt
 - Exhibit B – Pactiv GRAS Notice
- Exhibit 8 – KSR International Co., Petition v. Teleflex Inc. et al.,
No. 04-1350, Supreme Court of the United States
- Exhibit 9 – Leapfrog Enterprises, Inc. v. Fisher-Price, Inc. and Mattel, Inc.,
No. 06-1402, United States Court of Appeals for the Federal Circuit
- Exhibit 10 – “The Storage Life Of Beef And Pork Packaged In An Atmosphere With
Low Carbon Monoxide And High Carbon Dioxide” by Sorheim et al.
- Exhibit 11 – 1962 letters between FDA and Whirlpool Corporation
- Exhibit 12 – FDA Petition by Kalsec, Inc.
- Exhibit 13 – Color Atlas & Textbook of Hematology, Wm Platt, 2nd edition 1979
- Exhibit 14 – 21 U.S.C. § 121.1060 dated August 2, 1961
- Exhibit 15 – 21 U.S.C. § 121.1060 dated December 14, 1962
- Exhibit 16 – 21 C.F.R. § 173.350
- Exhibit 17 – FDA No Questions Letter from 2002/2003 to Pactiv from Eric Greenberg



PATENT

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Appl. No. : 09/915,150
Applicant : Gary R. DelDuca *et al.*
Filed : July 25, 2001
Title : Modified Atmospheric Packages and Methods for Making the Same

TC/A.U. : 1761
Examiner : Robert A. Madsen

Docket No. : 47097-01080

DECLARATION OF GARY R. DELDUCA
UNDER 37 C.F.R. § 1.132

Mail Stop Amendments
Commissioner for Patents
P.O. Box 1450
Alexandria, Virginia 22313

CERTIFICATE OF MAILING
37 C.F.R. 1.8

I hereby certify that this correspondence is being deposited with the U.S. Postal Service as First Class Mail in an envelope addressed to: Mail Stop Amendments, Commissioner for Patents, P.O. Box 1450, Alexandria, Virginia 22313 on the date indicated below:

September 8, 2003
Date

Admonne White
Admonne White

Dear Commissioner:

I, Gary R. DelDuca, declare that:

1. I hold a degree of B.S. in Mechanical Engineering From Rochester Institute of Technology in Rochester, New York that was obtained in 1980.
2. From 1980-1995, I worked as a developmental and senior engineer for Mobil Chemical Company, Plastics Division. As a developmental engineer, I worked in process and product development in the area of foam products. As a senior engineer, some of my responsibilities included designing specialized machinery that included machinery directed to stacking trays for meat processes. Mobil Chemical Company, Plastics Division was purchased by Tenneco Inc. in 1995. From 1995 to the present, I have been a Technical Manager for Tenneco Packaging Inc. in the area of modified atmosphere packaging (MAP) for meats. My responsibilities have included designing, developing, and implementing such modified

atmosphere packaging for meat and processes using the same. In 1999, Tenneco Packaging Inc. was renamed Pactiv Corporation.

4. The present invention is directed to methods of manufacturing a modified atmosphere package that includes carbon monoxide (CO). The invention has several advantages: (a) the "seasoning" period of the raw meat may be reduced or eliminated; (b) the ability to obtain consistent blooming with cuts off pigment-sensitive meats (*e.g.*, round bone) is improved; and (c) the ability to avoid "fixing" the color of the meat pigment to red. *See, e.g.*, page 11, line 29 - page 12, line 15; page 13, lines 11-17 of the application.

5. The "seasoning" period is the time period needed to diffuse the oxygen so that the meat has the ability to fully bloom. Page 3, lines 17-19 of the application. Trays, such as polystyrene foam trays, have a substantial amount of oxygen contained in its cellular structure that results in a time period of as long as about 5 to about 6 days to diffuse the oxygen contained in its cellular structure. Page 3, lines 21-23 of the application. If a foam tray is not used, the "seasoning" period can be reduced to one or two days. Page 3, lines 24-25 of the application. The reduction or elimination of the seasoning period "allows the meat to be displayed for retail sale much sooner than in existing low oxygen packaging systems." Page 11, line 32 - page 12, line 2 of the application. Seasoning periods are not desired by the retailers or packers because of the "need to store and maintain the meat-filled packages for an extended duration before being opened for retail sale." Page 3, lines 27-28 of the application.

6. Importantly, the present invention does not "fix" the color of the meat pigment to red with its use of CO, but rather the meat pigment tends to turn brown in a natural time period. *See* page 12, lines 10-12 of the application.

7. It is important to prevent the meat color from being "fixed" because it is unsafe (and potentially dangerous) to consume a piece of meat that has a bright red color that consumers associate with freshness, but has an unacceptable amount of bacteria. The present invention "surprisingly allows the meat pigment to convert to metmyoglobin in a similar fashion as fresh, raw meat in a retail environment." Page 12, lines 7-10 of the application. Specifically, the color of the meat after exposure to the ambient atmosphere degrades in a fashion not beyond the point of microbial soundness as if the CO had never been added to the modified packaging system.

8. The meat used in the modified atmosphere packaging of the present invention substantially maintains its color during the shipping process because the package has a modified atmosphere that includes from about 0.1% to about 0.8% CO. In one method, after removal of the package that is substantially permeable to oxygen, the CO is lost to the atmosphere. See page 12, lines 2-6 of the application. The CO may be lost to the atmosphere through the package that includes a non-barrier portion that is substantially permeable to oxygen. See *id.* and page 13, lines 5-10 of the application. This allows the conversion of the carboxymyoglobin to oxymyoglobin by using the oxygen from the air. Page 12, lines 4-7 of the application. The "gas mixture used in the modified atmosphere packages of the present invention, after removal, allows the carboxymyoglobin to convert to oxymyoglobin and then to metmyoglobin (brown) in a natural time period." Thus, the present invention does not "fix" the color. Page 12, lines 3-5 of the application.

9. Carbon monoxide (CO) has not been allowed to be used with fresh meat in the United States for about 40 years. The Food and Drug Administration ("FDA") regulation that currently prevents using CO with meat packaging systems in the United States is 21 C.F.R. § 173.350.

10. The concern of the FDA is believed to be that CO fixes the fresh meat color to a degree that allows the retailer to sell meat that looks good (a bright red color), but is unsafe and potentially dangerous to consume because it has unacceptable levels of bacteria. This act of fixing the meat color to a bright red color is referred to as "economic adulteration."

11. After about 40 years of not allowing CO to be used with fresh meats in the United States, the Applicants came up with novel approaches of using CO in modified atmosphere packaging (MAP) systems that avoided the concerns of "fixing" the meat color.

12. Pactiv Corporation, the assignee of the present invention, then gave notice to the FDA of a specific embodiment and process and evidence supporting Pactiv's conclusion that CO as used is GRAS (generally recognized as safe). The MAP system in Pactiv's GRAS notice may be used for packaging meats such as fresh cuts of case-ready muscle meat and ground case-ready meat to maintain wholesomeness, provide flexibility in distribution, and losses due to spoilage at retail sale.


13. The specific MAP system that was presented in the GRAS notice used 0.4% CO in a meat packaging system and was referred to as Pactiv's ActiveTech™ meat packaging system. The ActiveTech™ meat packaging system traditionally includes meats being placed in polystyrene trays and covered with oxygen-permeable, polyvinyl chloride ("PVC") overwraps. The wrapped trays of meat are then placed in an outer barrier bag. Ambient air is removed and replaced with a blend of 0.4% CO, 30% carbon dioxide, and the balance being nitrogen. The myoglobin of the meat converts from oxymyoglobin to carboxymyoglobin (red). The meat maintains its red color while in storage until the package is opened for retail display by removing the outer barrier bag. The package will lose CO to the atmosphere and, thus, the retail display will not have CO. Once in retail display, the meat's myoglobin begins its natural conversion to metmyoglobin (brown). The CO used in the Pactiv MAP system did not mask the spoilage or extend the color life beyond the point of wholesomeness (*i.e.*, the point of microbial soundness).

14. The FDA stated that it had no questions regarding Pactiv's conclusion about Pactiv's ActiveTech™ meat packaging system using 0.4% CO being GRAS because of the evidence presented by Pactiv in its notice. This FDA review allows Pactiv to use CO with fresh meat in its application. It is believed to be the first system to overcome the prohibition of CO with fresh meat in the U.S. in the last 40 years.

15 I hereby declare that all statements made of my own knowledge are true and that all statements made on information and belief are believed to be true; and, further, that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment or both under Section 1001 of Title 18 of the United States Code, and that such willful false statements may jeopardize the validity of the application or any patent issuing thereon.

Date:

9/8/03


Gary R. DelDuca



PATENT

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Appl. No. : 09/915,150
Applicant : Gary R. DelDuca *et al.*
Filed : July 25, 2001
Title : Modified Atmospheric Packages and Methods for Making the Same

TC/A.U. : 1761
Examiner : Robert A. Madsen

Docket No. : 47097-01080

SECOND DECLARATION OF GARY R. DELDUCA
UNDER 37 C.F.R. § 1.132

Mail Stop Amendments
Commissioner for Patents
P.O. Box 1450
Alexandria, Virginia 22313

CERTIFICATE OF MAILING
37 C.F.R. 1.8

I hereby certify that this correspondence is being deposited with the U.S. Postal Service as First Class Mail in an envelope addressed to: Mail Stop Amendments, Commissioner for Patents, P.O. Box 1450, Alexandria, Virginia 22313 on the date indicated below:

June 16, 2004
Date


Adrienne White

Dear Commissioner:

I, Gary R. DelDuca, declare that:

1. I hold a degree of B.S. in Mechanical Engineering From Rochester Institute of Technology in Rochester, New York that was obtained in 1980.
2. From 1980-1995, I worked as a developmental and senior engineer for Mobil Chemical Company, Plastics Division. As a developmental engineer, I worked in process and product development in the area of foam products. As a senior engineer, some of my responsibilities included designing specialized machinery that included machinery directed to stacking trays for meat processes. Mobil Chemical Company, Plastics Division was purchased by Tenneco Inc. in 1995. From 1995 to the present, I have been a Technical Manager for

Tenneco Packaging Inc. in the area of modified atmosphere packaging (MAP) for meats. My responsibilities have included designing, developing, and implementing such modified atmosphere packaging for meat and processes using the same. In 1999, Tenneco Packaging Inc. was renamed Pactiv Corporation ("Pactiv").

3. I am familiar with claims 1-37, 87-90, and 161-171, that are directed to methods of manufacturing a modified atmosphere package. I am aware of the Office Action dated February 17, 2004, and the obviousness rejections in that Office Action. I understand that the analysis of the patentability of claims 1-37, 87-90 and 161-171 should take into account certain facts related to the commercial success, and the clinical or verification success of the MAP method that is covered by these claims. I wish to provide evidence showing that the Pactiv improved ActiveTech® meat packaging system and process have been commercially successful. Additionally, I wish to provide evidence that the Pactiv improved ActiveTech® meat packaging system and process have achieved substantial clinical effectiveness.

4. Pactiv and its predecessor Tenneco Packaging Inc.¹ have sold modified atmosphere packaging systems beginning in 1998 (the traditional ActiveTech® meat packaging system). The Pactiv traditional ActiveTech® meat packaging system includes meats being placed in polystyrene trays and covered with oxygen-permeable, polyvinyl chloride ("PVC") overwraps. The wrapped trays of meat are then placed in an outer barrier bag. Ambient air is removed and replaced with a blend of 30 vol.% carbon dioxide, and the balance being nitrogen.

5. Beginning in March of 2002, Pactiv began offering for sale an improved ActiveTech® meat packaging system. Pactiv's improved ActiveTech® meat packaging system

¹ These will be collectively referred to Pactiv Corporation in the remainder of the declaration.

includes meats being placed in polystyrene trays and covered with oxygen-permeable, PVC overwraps. The wrapped trays of meat are then placed in an outer barrier bag. Ambient air is removed and replaced with a blend of 0.4 vol.% carbon monoxide (CO), 30 vol.% carbon dioxide, and the balance being nitrogen.

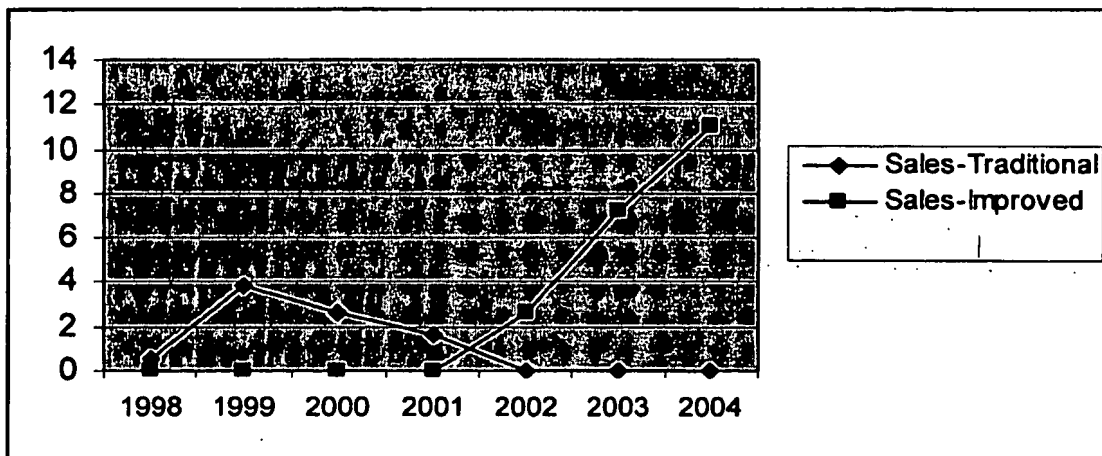
6. The modified atmosphere used in Pactiv's improved ActiveTech® meat packaging system differs from the modified atmosphere used in the Pactiv's traditional ActiveTech® meat packaging system. Specifically, Pactiv's improved ActiveTech® meat packaging system uses 0.4 vol.% CO, while Pactiv's traditional ActiveTech® meat packaging system does not use CO. Because of the addition of CO, the equipment used in Pactiv's improved ActiveTech® meat packaging system may vary slightly as compared to Pactiv's traditional ActiveTech® meat packaging system. Specifically, a mixer may be added to Pactiv's improved ActiveTech® meat packaging system to mix the CO, carbon dioxide, and nitrogen. Additionally, a CO gas recovery hood and safety features may also be included in Pactiv's improved ActiveTech® meat packaging system.

7. The purchasers of either Pactiv's improved ActiveTech® meat packaging system or Pactiv's traditional ActiveTech® meat packaging system receive a license for the process and the knowledge to run such a process. Pactiv allows its customers to use its oxygen-absorber dispensing-machine at no cost. The remaining machinery used to perform either Pactiv's improved ActiveTech® meat packaging system or Pactiv's traditional ActiveTech® meat packaging system is purchased by the customer. Typically, this remaining machinery is sold by

Pactiv to its customers. The customers also typically purchase the oxygen absorbers, trays, and film from Pactiv.

8. As shown below in the Graph and the Table, sales of Pactiv's traditional ActiveTech® meat packaging system were decreasing in 2000 and 2001. The sales of Pactiv's improved ActiveTech® meat packaging system, however, have increased at an exponential rate since its introduction in March of 2002. The sales of Pactiv's improved ActiveTech® meat packaging system have been commercially successful with sales numbers of over 7 million in 2003 and an estimated sales number of 11 million in 2004. The sales numbers below include the total of the purchased licenses, the purchased remaining machinery, and supplies (which include oxygen absorbers, activator fluid, and film).

GRAPH



TABLE

U.S. Sales Year	Sales of Traditional ActiveTech® (in millions)	Sales of Improved ActiveTech® (in millions) ²
1998	0.5	0
1999	3.8	0
2000	2.6	0
2001	1.6	0
2002	0	2.8
2003	0	7.2
2004	0	11 ³

9. Since March of 2002, both Pactiv's improved ActiveTech® meat packaging system and Pactiv's traditional ActiveTech® meat packaging system have been available for sale. Since March 2002, no customer has purchased Pactiv's traditional ActiveTech® meat packaging system. In fact, every customer still practicing Pactiv's technology has converted its traditional ActiveTech® meat packaging system into Pactiv's improved ActiveTech® meat packaging system. Thus, to my knowledge no customer is still practicing Pactiv's traditional ActiveTech® meat packaging system. It can be concluded that these customers prefer the Pactiv's improved ActiveTech® meat packaging system over Pactiv's traditional ActiveTech® meat packaging system. The cost of Pactiv's improved ActiveTech® meat packaging system versus Pactiv's traditional ActiveTech® meat packaging system is fractionally more expensive. Thus, the commercial success of Pactiv's improved ActiveTech® meat packaging system cannot be attributed to a cost advantage.

² Pactiv's improved ActiveTech™ meat packaging system was not offered for sale until March 2002.

³ This is an estimated figure based on sales through April of 2004.

10. The process of manufacturing using Pactiv's improved ActiveTech® meat packaging system is an example of a process that would be covered under independent claims 1, 22 and 161 of the present application.

11. After about 40 years of not allowing CO to be used with fresh meats in the United States, the Applicants came up with novel approaches of using CO in modified atmosphere packaging (MAP) systems that avoided the concerns of "fixing" the meat color, which can mask the spoilage or extend the life beyond the point of microbial soundness. The problem of fixing color using CO is known to those skilled in the art. One example of a reference that recognizes this problem is an article entitled "The storage life of beef and pork packaged in an atmosphere with low carbon monoxide and high carbon monoxide" to Sorheim, Nissen and Nesbakken. This article was discussed in the Office Action dated May 7, 2003.

12. The FDA stated that it had no questions regarding Pactiv's conclusion about Pactiv's improved ActiveTech® meat packaging system using 0.4% CO being GRAS because of the evidence presented by Pactiv in its GRAS notice. This FDA review allows Pactiv to use CO with fresh meat in its application. It is believed to be the first system to overcome the prohibition of CO with fresh meat in the U.S. in the last 40 years. Thus, an important advancement in the art of meat packaging systems has been accomplished by the present invention. The importance has been recognized by the customers of Pactiv's improved ActiveTech® meat packaging system and process.

13. I hereby declare that all statements made of my own knowledge are true and that all statements made on information and belief are believed to be true; and, further, that these statements were made with the knowledge that willful false statements and the like so made are

punishable by fine or imprisonment or both under Section 1001 of Title 18 of the United States Code, and that such willful false statements may jeopardize the validity of the application or any patent issuing thereon.

Date: June 16, 2004


Gary R. DelDuca



PATENT

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Appl. No. : 09/915,150
Applicant : Gary R. DelDuca *et al.*
Filed : July 25, 2001
Title : Modified Atmospheric Packages and Methods for Making the Same

TC/A.U. : 1761
Examiner : Robert A. Madsen

Docket No. : 47097-01080

THIRD DECLARATION OF GARY R. DELDUCA
UNDER 37 C.F.R. § 1.132

Mail Stop Amendments
Commissioner for Patents
P.O. Box 1450
Alexandria, Virginia 22313

CERTIFICATE OF MAILING
37 C.F.R. 1.8

I hereby certify that this correspondence is being deposited with the U.S. Postal Service as First Class Mail in an envelope addressed to: Mail Stop Amendments, Commissioner for Patents, P.O. Box 1450, Alexandria, Virginia 22313 on the date indicated below:

5/10/05
Date

Adrienne White
Adrienne White

Dear Commissioner:

I, Gary R. DelDuca, declare that:

1. I hold a degree of B.S. in Mechanical Engineering From Rochester Institute of Technology in Rochester, New York that was obtained in 1980.

2. From 1980-1995, I worked as a developmental and senior engineer for Mobil Chemical Company, Plastics Division. As a developmental engineer, I worked in process and product development in the area of foam products. As a senior engineer, some of my responsibilities included designing specialized machinery that included machinery directed to stacking trays for meat processes. Mobil Chemical Company, Plastics Division was purchased by Tenneco Inc. in 1995. From 1995 to the present, I have been a Technical Manager for Tenneco Packaging Inc. in the area of modified atmosphere packaging (MAP) for meats. My responsibilities have included designing, developing, and implementing such modified atmosphere

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packaging for meat and processes using the same. In 1999, Tenneco Packaging Inc. was renamed Pactiv Corporation ("Pactiv").

3. One important aspect of the present invention is that the present invention does not "fix" the color of the meat pigment to red with its use of carbon monoxide (CO), but rather the meat pigment tends to turn brown in a natural time period after removal of the second package that is substantially impermeable to oxygen. It is important to prevent the meat color from being "fixed" because it is unsafe (and potentially dangerous) to consume a piece of meat that has a bright red color that consumers associate with freshness, but is beyond the point of microbial soundness. The term "fix" in this context does not mean that the color of meat pigment never changes to a brown color, but rather that the meat pigment does not turn brown in a natural time period after the meat pigment is exposed to the atmosphere.

4. It is known to those skilled in the art that when hemoglobin in the red blood cells is exposed to CO, it has a much greater affinity than oxygen does with hemoglobin. In fact, when hemoglobin in the red blood cells is exposed to CO, the CO has an affinity 200 times greater than oxygen with hemoglobin. Therefore, one skilled in the art would expect that CO "fixes" the color of the meat pigment past its natural time period upon exposure to the atmosphere. In other words, because of the hemoglobin's high affinity towards CO, the pigment of the meat, prior to Applicants' invention, would not have been expected to degrade in a natural time period.

5. U.S. Patent No. 3,459,117 to Koch discloses (a) covering primal cuts made at a slaughterhouse with a film that contains a small quantity of CO, (b) removing the CO-containing film at the retail outlet, and (c) cutting the primal cuts into individual steaks, roasts, etc.

6. "Primal" cuts of meat at the time of the Koch disclosure (late 1960's), however, generally refers to sections of meat from anywhere between about 50 and 150 or more lbs. The term "subprimal" cuts of meat is used today and generally refers to cuts of meat from about 15 to about 20 lbs. Thus, it is clear that the term primal cuts of meat in Koch refers to a large quantity of meat.

7. It would not be reasonable to one of ordinary skill in the art that a 50-150 lb piece of meat disclosed in Koch that had been exposed to a small quantity of CO would turn the non-surface meat pigments, which were not exposed to CO, to carboxymyoglobin. This is supported by the disclosure of U.S. Patent No. 6,042,859 to Shalvai. The disclosure of Shalvai with 100%

CO (as compared to the small quantity of CO in Koch) took over 7 days to saturate a small piece of meat with CO. Specifically, in Example 3 of Shaktai, 0.5 to 1.5Kg (about 1.4 lbs to about 4.2 lbs) took 7 days upon exposure to 100% CO to turn the meat pigment to carboxymyoglobin. Therefore, when the primal cuts of meat of Koch were cut at the retail outlet into individual steaks and roasts, the meat pigments of such individual steaks and roasts had not been exposed to the CO from the CO-containing film.

8. It would be expected that the individually cut steaks and roasts sections of Koch that were not exposed to CO would degrade in a manner similar to other similar cuts of steaks and roasts that had also not been exposed to CO. Thus, Koch teaches that meat pigment in the form of individual steaks and roasts not exposed to CO in the CO-containing film would degrade in a similar manner of steaks and roasts not treated with CO. Thus, Koch does not teach or suggest that the use of CO turns meat pigments brown in a natural time period after removal of the CO-containing film.

9. I hereby declare that all statements made of my own knowledge are true and that all statements made on information and belief are believed to be true; and, further, that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment or both under Section 1001 of Title 18 of the United States Code, and that such willful false statements may jeopardize the validity of the application or any patent issuing thereon.

Date: May 9, 2005


Gary R. DelDuca



PATENT

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Appl. No. : 09/915,150
Applicant : Gary R. DelDuca *et al.*
Filed : July 25, 2001
Title : Methods for Making Modified Atmospheric Packages

TC/A.U. : 1761
Examiner : Robert A. Madsen

Docket No. : 47097-01080

FOURTH DECLARATION OF GARY R. DELDUCA
UNDER 37 C.F.R. § 1.132

Mail Stop Amendment
Commissioner for Patents
P.O. Box 1450
Alexandria, Virginia 22313

CERTIFICATE OF MAILING
37 C.F.R. 1.8

I hereby certify that this correspondence is being deposited with the U.S. Postal Service as First Class Mail in an envelope addressed to: Mail Stop Amendment, Commissioner for Patents, P.O. Box 1450, Alexandria, Virginia 22313 on the date indicated below:

11/2/2005
Date


Adrienne White

Dear Commissioner:

I, Gary R. DelDuca, declare that:

1. I hold a degree of B.S. in Mechanical Engineering From Rochester Institute of Technology in Rochester, New York that was obtained in 1980.
2. From 1980-1995, I worked as a developmental and senior engineer for Mobil Chemical Company, Plastics Division. As a developmental engineer, I worked in process and product development in the area of foam products. As a senior engineer, some of my responsibilities included designing specialized machinery that included machinery directed to stacking trays for meat processes. Mobil Chemical Company, Plastics Division was purchased by Tenneco Inc. in 1995. From 1995 to the present, I have been a Technical Manager for

Tenneco Packaging Inc. in the area of modified atmosphere packaging (MAP) for meats. My responsibilities have included designing, developing, and implementing such modified atmosphere packaging for meat and processes using the same. In 1999, Tenneco Packaging Inc. was renamed Pactiv Corporation ("Pactiv").

3. I am aware of the Office Action dated August 2, 2005, and have read the portion of the Office Action discussing the phrase "turns brown in a natural time period." This phrase is used in independent claims 1, 22 and 161 and disclosed in the patent application at, for example, page 11, line 29 – page 12, line 15.

4. The phrase "turns brown in a natural time period" is a phrase that is used and understood by those skilled in the art. This phrase has been used in correspondence related to meat-packaging systems between retailers and myself. Specifically, this phrase has been used by those skilled in the art in the context of the color of the meat pigment. It is important to retailers and food packers that the color of the meat pigment not be fixed and turns brown in a natural time period.

5. One example of this phrase being used in the published literature is shown in Exhibit A (Principles and Applications of Modified Atmosphere Packaging of Food). On page 283, the literature discusses the effect of the meat turning brown in connection with conventionally overwrapped trays and also discusses that the color stability is limited on the shelf-life depending on type of meat (muscle).

6. The portion "turns brown" of the phrase "turns brown in a natural time period" means that the piece of meat has some brown, but does not mean that the piece of meat has to be 100% brown. Retailers and food packers use the phrase "turns brown" in the context of whether

most customers would consider the color of the meat pigment undesirable such that the customers would not purchase the meat. The phrase "turns brown" is frequently used by retailers and food packers and, thus, is not indefinite.

7. The term "natural time period" of the phrase "turns brown in a natural time period" cannot be uniquely defined because the color of the meat pigment varies between the type of meat and the conditions for displaying such meat. See page 20, lines 17-26 of the present application ("The display times varied based on product type, initial microbial loads and storage conditions."). The natural time period for the meat pigment turning brown is not the same between ground beef, strip loins (strip steaks), inside portion of inside round steaks, outer portion of inside rounds steaks, and tenderloins. For example, the natural time period in which the meat pigment turns brown is about 4 days for strip steaks, while the natural time period in which the meat pigment turns brown for tenderloin is about 1 day.

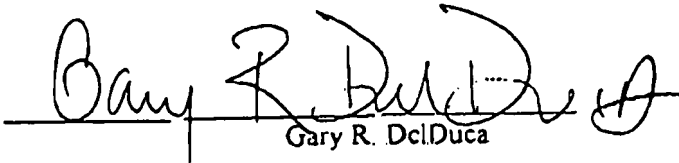
8. I am not aware of any standard test for determining the color of the meat pigment. The most common type of testing for determining the color of meat pigment is a visual inspection to determine whether the color of the meat pigment is acceptable for sale. As discussed in the patent application, the color of the meat pigment can be visually determined. Page 20, line 27 – page 21, line 6 of the present application. In the examples of the present application, the color of the meat pigment was visually determined using a five-point scale where 1 = very bright red, 2 = bright red, 3 = slightly dark red or tan, 4 = moderately dark red or tan, and 5 = extremely dark red or brown. Page 20, lines 28-30 of the present application. If the score was 3.5 or less, then it was visually determined that the meat pigment was an acceptable color. Page 20, lines 30-31 of the present application.

9. Alternatively, there are other tests that are used to determine the redness of the meat pigment. One example of a test for redness was disclosed in the present application at page 21, lines 7-16. In this test, examples were instrumentally analyzed for redness (a^*) using a colorimeter or photometer. See page 21, lines 8-11 of the present application. Normally, a^* values (higher values indicate more redness) are highly correlated to visual appraisal. Page 21, lines 12-13 of the present application. This type of test is not more accurate than a visual inspection by those skilled in the art because the color of the meat pigment does not degrade in a uniform fashion. Thus, some portions of the meat pigment may be brown and other portions of the meat pigment may be red, which may make the a^* test less accurate than visual inspection.

10. In summary, the phrase "turns brown in a natural time period" as used in the context of independent claims 1, 22 and 161 is understood by those skilled in the art and is not an indefinite phrase in this context.

11. I hereby declare that all statements made of my own knowledge are true and that all statements made on information and belief are believed to be true; and, further, that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment or both under Section 1001 of Title 18 of the United States Code, and that such willful false statements may jeopardize the validity of the application or any patent issuing thereon.

Date: November 2, 2005


Gary R. DeDuca

PRINCIPLES AND APPLICATIONS OF MODIFIED ATMOSPHERE PACKAGING OF FOOD

Edited by R. T. Parry



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may be vacuum packaged, but it is difficult to remove all the air from the system. Flushing with carbon dioxide removes residual oxygen from the system. Recently, Garout *et al.* (1989) reported an increased storage life for lamb loins and carcasses, packed in carbon dioxide, compared with similar vacuum-packaged meat in consignments transported by air from New Zealand to Saudi Arabia. Given similar chilling conditions, the storage life of CO₂-packed lamb was about 40 days longer than that of vacuum-packed lamb. Storage life was limited by the development of putrid spoilage, principally due to psychrotrophic enterobacteria.

11.6 Retail marketing

11.6.1 Consumer cuts

The universal preference for bright-red colour in fresh meat is a major factor in determining the way fresh red meat is packaged for retail sale. This preference is strong in the case of beef and lamb, both of which have a relatively high pigment content, but is less important in pork and veal with their much paler colour. At the point of sale, colour and colour stability are the most important attributes of meat quality and various ways have been used to fulfil consumer expectation that an attractive bright-red colour is compatible with long shelf-life and good eating quality.

There are three types of packaging method suitable for the presentation and display of consumer joints and cuts of meat. These are (i) conventional aerobic overwrapped trays; (ii) MAP, especially using higher levels of oxygen; and (iii) vacuum-packaging. All have been used in the retail market-place to varying extent. The use of vacuum-packaging in retail marketing is limited, due to its purple colour, and attempts to educate consumers to accept the colour on the basis that there is a greatly extended shelf-life have been largely unsuccessful. Nevertheless there is a limited specialised market for the product and interest in the system remains for some marketing situations.

11.6.2 Conventionally overwrapped trays

The conventional aerobic method of packaging widely used by supermarkets and other self-service outlets for retail presentation, involves placing the meat in semi-rigid plastic trays, which are then overwrapped with a clear, gas permeable plastic film which readily allows an unrestricted supply of oxygen to the pigment. The film is usually a light-gauge vinyl or polyethylene derivative which combines a low permeability to water vapour and a high permeability to oxygen ($>10\,000\text{ cm}^3\text{ m}^{-2}\text{ day}^{-1}\text{ atm}^{-1}\text{ O}_2$). A wide range of films is currently available commercially for overwrapping trays of meat. All have adequate oxygen permeability and the choice among them is

more likely to be based on price, suitability for use on machines, optical clarity and sealability (Taylor, 1985).

Under the aerobic conditions prevailing in an overwrapped tray, pseudo-mounds grow readily, and this will lead to relatively rapid spoilage of consumer cuts on retail display. More usually however, the limit to storage life is a result of biochemical discoloration, due to intrinsic enzymic action and oxidation. High bacterial contamination causes an accelerated deterioration in colour, due, initially at least, to competition for available oxygen. A reduction in the partial pressure of oxygen to the critical level for oxidation to metmyoglobin results in the meat turning brown. Development of browning causes 'fading' and a 'tired' appearance of the packaged meat, limiting its display life to a maximum of two to three days. Hood and Kiordan (1973) reported that when packs of discoloured meat sold from a retail display cabinet are compared with similar packs of bright-red meat, there is a considerable bias against the sale of the discoloured meat, equivalent to a ratio of 2:1 when the level of discoloration reaches 20% metmyoglobin. Under normal commercial display, McDougall (1972) found that colour stability limited effective shelf-life to two days depending on the muscle, before oxidising significantly to the unattractive brown metmyoglobin pigment. In practice, supermarkets generally restock every day to ensure the meat on retail display has a fresh appearance. A practical problem is the poor control of temperature that is often achieved in retail display cabinets (Taylor, 1982).

Taylor (1985) points out that overwrapped trays provide an effective thermal insulation so that the meat temperature can be higher than the surroundings. The lighting system in display cabinets can also produce a 'greenhouse' within the pack, heating the exposed surface. The meat must be adequately cooled before packing and careful control of temperature must be maintained not only in the cabinet but also within the package during storage and display. Temperature of storage and display is critical in obtaining maximum shelf-life. At low refrigeration temperature, an increase of 5°C can halve the colour shelf-life, depending on species and muscle. Low-temperature storage, as close as possible to 0°C, is essential to obtain maximum shelf-life. Even at this low temperature the maximum for beef fillet is only about four days (Hood, 1984). Whilst temperature is the single most important factor under practical commercial conditions other factors should also be taken into account. Muscle variability, UV light and length of time post-mortem are among the most important. Some cuts and especially certain muscles have a very short shelf-life with respect to colour stability even under ideal storage conditions, whilst others are significantly better in this respect. Thus, beef from the fillet (*M. psoas major*) has a colour shelf-life of 1-5 days at 5°C, whilst loin steak (*M. longissimus dorsi*) retains a bright-red colour for more than six days at the same temperature (O'Keefe and Hood, 1982).



PATENT

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

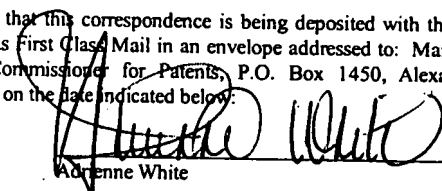
Appl. No. : 09/915,150
Applicant : Gary R. DelDuca *et al.*
Filed : July 25, 2001
Title : Methods for Making Modified Atmospheric Packages

TC/A.U. : 1761
Examiner : Robert A. Madsen

Docket No. : 47097-01080

**FIFTH DECLARATION OF GARY R. DELDUCA
UNDER 37 C.F.R. § 1.132**

Mail Stop Amendment
Commissioner for Patents
P.O. Box 1450
Alexandria, Virginia 22313

CERTIFICATE OF MAILING 37 C.F.R. 1.8	
I hereby certify that this correspondence is being deposited with the U.S. Postal Service as First Class Mail in an envelope addressed to: Mail Stop Amendment, Commissioner for Patents, P.O. Box 1450, Alexandria, Virginia 22313 on the date indicated below:	
05/25/2006 Date	 Adrienne White

Dear Commissioner:

I, Gary R. DelDuca, declare that:

1. I hold a degree of B.S. in Mechanical Engineering From Rochester Institute of Technology in Rochester, New York that was obtained in 1980.
2. From 1980-1995, I worked as a developmental and senior engineer for Mobil Chemical Company, Plastics Division. As a developmental engineer, I worked in process and product development in the area of foam products. As a senior engineer, some of my responsibilities included designing specialized machinery that included machinery directed to stacking trays for meat processes. Mobil Chemical Company, Plastics Division was purchased by Tenneco Inc. in 1995. From 1995 to the present, I have been a Technical Manager for Tenneco Packaging Inc. in the area of modified atmosphere packaging (MAP) for meats. My responsibilities have included designing, developing, and implementing such modified

atmosphere packaging for meat and processes using the same. In 1999, Tenneco Packaging Inc. was renamed Pactiv Corporation ("Pactiv").

3. In the Office Action dated January 25, 2006, it is stated that Woodruff "teaches surface contact of a meat with CO to maintain a red color, will have CO removably associated with the meat". Page 5.

4. Woodruff does not teach or suggest that the color of the meat pigment turns brown in a natural time period. For example, Woodruff in Example 1 discloses a 0.5 lb. beefsteak that was exposed to 0.5% CO, which was nearly all absorbed two days later. *See* col. 4, lines 34-48. After being exposed in a modified atmosphere that included 16% oxygen, "the beefsteak retained its good red color, and the carboxymyoglobin color had penetrated no more deeply than it had at the end of the two days." *See* Col. 4, lines 49-54. This passage implies that the carboxymyoglobin color was still retained within the beefsteak after 6 days despite being exposed to an atmosphere with a generally similar amount of oxygen as in air (compare about 21% oxygen to 16% oxygen). It would be expected to one skilled in the art that the beefsteak would turn brown in about 2-3 days, depending on the cut of meat. Thus, this example clearly shows that the beefsteak of Woodruff in Example 1 did not turn brown in a natural time period, but rather "fixed" the color of the meat pigment. Similarly, in Example 1 of Woodruff, a 0.5 lb. beefsteak exposed to 2.5% CO under similar conditions also retained its good color after 6 days. *See* col. 4, line 55- col. 5, line 6.

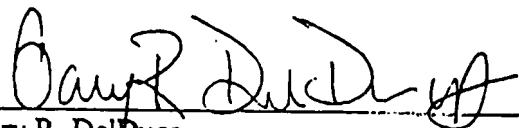
5. None of the other examples of Woodruff supports a modified atmosphere package wherein the CO associated with the raw meat is adapted to be removed such that the color of the meat pigment is not fixed and turns brown in a natural time period. Rather, the other examples of Woodruff generally disclose the condition of the meat pigment while being stored in a modified atmosphere containing CO.

6. In summary, Woodruff does not disclose, teach or suggest that the use of CO on meat pigment turns brown in a natural time period, but rather Woodruff teaches and suggests "fixing" the color of the meat pigment in Example 1.

7. Koch discloses that "[o]f course, if desired, the final cuts rather than just the primal cuts may be individually wrapped in the cover such as shown in FIGS. 1 and 2, this cover preferably being replaced with a conventional cover by the retailer." Col. 3, lines 13-16. This passage, however, does not disclose, teach or suggest that the color of the meat pigment is not fixed and will turn brown in a natural time period. Furthermore, this passage has nothing to do with the statement in the Office Action directed to Koch on the meat color ("Koch et al. teach a meat surface that has been exposed to CO for 7 days during storage under a modified atmosphere will remain red in color for 3 days after being removed from the modified atmosphere package[] and packaged in conventional wrapper at[] the retail outlet"). See page 5 of the Office Action. Rather, Koch discloses "[w]hen the primal cuts arrive at the retail outlet, the covers are removed and the meat is cut into individual steaks, roasts, etc. which may be separately wrapped in conventional wrapping materials. It has been found that meat will release a saleable red color for as long as 10 days when covered with the cover herein described for the first seven days and with a conventional cover for the remaining days." Col. 3, lines 5-13 of Koch (underlining added).

8. I hereby declare that all statements made of my own knowledge are true and that all statements made on information and belief are believed to be true; and, further, that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment or both under Section 1001 of Title 18 of the United States Code, and that such willful false statements may jeopardize the validity of the application or any patent issuing thereon.

Date: May 24, 2006



Gary R. DelDuca



PATENT

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Appl. No. : 09/915,150
Applicant : Gary R. DelDuca *et al.*
Filed : July 25, 2001
Title : Modified Atmospheric Packages and Methods for Making the Same

TC/A.U. : 1761
Examiner : Jyoti Chawla

Docket No. : 47097-01080

**SIXTH DECLARATION OF GARY R. DELDUCA
UNDER 37 C.F.R. § 1.132**

Mail Stop Amendment
Commissioner for Patents
P.O. Box 1450
Alexandria, Virginia 22313

Dear Commissioner:

I, Gary R. DelDuca, declare that:

1. I hold a degree of B.S. in Mechanical Engineering From Rochester Institute of Technology in Rochester, New York that was obtained in 1980.
2. From 1980-1995, I worked as a developmental and senior engineer for Mobil Chemical Company, Plastics Division. As a developmental engineer, I worked in process and product development in the area of foam products. As a senior engineer, some of my responsibilities included designing specialized machinery that included machinery directed to stacking trays for meat processes. Mobil Chemical Company, Plastics Division was purchased by Tenneco Inc. in 1995. From 1995 to the present, I have been a Technical Manager and/or Technical Sales Manager for Tenneco Packaging Inc. in the area of modified atmosphere packaging (MAP) for meats. My responsibilities have included designing, developing, and

implementing such modified atmosphere packaging for meat and processes using the same. In 1999, Tenneco Packaging Inc. was renamed Pactiv Corporation ("Pactiv").

3. In The Office Action dated August 10, 2006, one of the arguments raised with respect to commercial success was that "it is not clear if the claimed invention resulted in the commercial success or whether other factors contributed to the success, such as increase[d] advertising/marketing." Page 10 of the Office Action. I wish to provide additional evidence showing that the Pactiv improved ActiveTech® meat packaging system and process have been commercially successful without increased advertising/marketing.

4. Pactiv and its predecessor Tenneco Packaging Inc.¹ have sold modified atmosphere packaging systems beginning in 1998 (the traditional ActiveTech® meat packaging system). The Pactiv traditional ActiveTech® meat packaging system includes meats being placed in polystyrene trays and covered with oxygen-permeable, polyvinyl chloride ("PVC") overwraps. The wrapped trays of meat are then placed in an outer barrier bag. Ambient air is removed and replaced with a blend of 30 vol.% carbon dioxide, and the balance being nitrogen.

5. Beginning in March of 2002, Pactiv began offering for sale an improved ActiveTech® meat packaging system. Pactiv's improved ActiveTech® meat packaging system includes meats being placed in polystyrene trays and covered with oxygen-permeable, PVC overwraps. The wrapped trays of meat are then placed in an outer barrier bag. Ambient air is removed and replaced with a blend of 0.4 vol.% carbon monoxide (CO), 30 vol.% carbon dioxide, and the balance being nitrogen.

6. The modified atmosphere used in Pactiv's improved ActiveTech® meat packaging system differs from the modified atmosphere used in the Pactiv's traditional ActiveTech® meat packaging system. Specifically, Pactiv's improved ActiveTech® meat packaging system uses 0.4 vol.% CO, while Pactiv's traditional ActiveTech® meat packaging system does not use CO. Because of the addition of CO, the equipment used in Pactiv's improved ActiveTech® meat packaging system may vary slightly as compared to Pactiv's traditional ActiveTech® meat packaging system. Specifically, a mixer may be added to Pactiv's improved ActiveTech® meat packaging system to mix the CO, carbon dioxide, and nitrogen.

¹ These will be collectively referred to Pactiv Corporation in the remainder of the declaration.

Additionally, a CO gas recovery hood and safety features may also be included in Pactiv's improved ActiveTech® meat packaging system.

7. The purchasers of either Pactiv's improved ActiveTech® meat packaging system or Pactiv's traditional ActiveTech® meat packaging system receive a license for the process and the knowledge to run such a process. Pactiv allows its customers to use its oxygen-absorber dispensing-machine at no cost. The remaining machinery used to perform either Pactiv's improved ActiveTech® meat packaging system or Pactiv's traditional ActiveTech® meat packaging system is purchased by the customer. Typically, this remaining machinery is sold by Pactiv to its customers. The customers also typically purchase the oxygen absorbers, trays, and film from Pactiv.

8. As shown below in the Table, sales of Pactiv's traditional ActiveTech® meat packaging system were decreasing in 2000 and 2001. The sales of Pactiv's improved ActiveTech® meat packaging system, however, have substantially increased since its introduction in March of 2002. The sales of Pactiv's improved ActiveTech® meat packaging system have been commercially successful with sales numbers of about or over 6 million in each of the years since 2003. The sales numbers below include the total of the purchased licenses, the purchased remaining machinery, and supplies (which include oxygen absorbers, activator fluid, and film).

TABLE

U.S. Sales Year	Sales of Traditional ActiveTech® (in millions)	Sales of Improved ActiveTech® (in millions)²
1998	0.5	0
1999	3.8	0
2000	2.6	0
2001	1.6	0
2002	0	2.8
2003	0	7.2
2004	0	7.1
2005	0	6.5
2006	0	4.5 ³

9. Since March of 2002, both Pactiv's improved ActiveTech® meat packaging system and Pactiv's traditional ActiveTech® meat packaging system have been available for sale. Since March 2002, no customer has purchased Pactiv's traditional ActiveTech® meat packaging system. In fact, every customer still practicing Pactiv's technology has converted its traditional ActiveTech® meat packaging system into Pactiv's improved ActiveTech® meat packaging system. Thus, to my knowledge no customer is still practicing Pactiv's traditional ActiveTech® meat packaging system. It can be concluded that these customers prefer the Pactiv's improved ActiveTech® meat packaging system over Pactiv's traditional ActiveTech® meat packaging system. The cost of Pactiv's improved ActiveTech® meat packaging system versus Pactiv's traditional ActiveTech® meat packaging system is fractionally more expensive. Thus, the commercial success of Pactiv's improved ActiveTech® meat packaging system cannot be attributed to a cost advantage.

² Pactiv's improved ActiveTech™ meat packaging system was not offered for sale until March 2002.


³ Sales through September of 2006.

10. Since 2002, there has been no increase in the number of sales personnel from Pactiv who are responsible for sales of Pactiv's improved ActiveTech® meat packaging system. In fact, the number of sales personnel who are responsible for sales of the Pactiv's improved ActiveTech® meat packaging system have decreased since 2002. There has been little or no advertising directed to sales of Pactiv's improved ActiveTech® meat packaging system since 2002. The amount of advertising, if any, has not increased since 2002 and likely has decreased substantially from that directed to Pactiv's traditional ActiveTech® meat packaging system. Thus, the commercial success of Pactiv's improved ActiveTech® meat packaging system cannot be attributed to increased marketing/advertising.

11. The process of manufacturing using Pactiv's improved ActiveTech® meat packaging system is an example of a process that would be covered under independent claims 1, 22 and 161 of the present application.

12. I hereby declare that all statements made of my own knowledge are true and that all statements made on information and belief are believed to be true; and, further, that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment or both under Section 1001 of Title 18 of the United States Code, and that such willful false statements may jeopardize the validity of the application or any patent issuing thereon.

Date: November 6, 2006



Gary R. DeDuca



PATENT

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Appl. No. : 09/915,150
Applicant : Gary R. DelDuca *et al.*
Filed : July 25, 2001
Title : Modified Atmospheric Packages and Methods for Making the Same

TC/A.U. : 1761
Examiner : Robert A. Madsen

Docket No. : 47097-01080

DECLARATION OF DR. MELVIN C. HUNT
UNDER 37 C.F.R. § 1.132

Mail Stop Amendment-Fee
Commissioner for Patents
P.O. Box 1450
Alexandria, Virginia 22313

CERTIFICATE OF MAILING
37 C.F.R. 1.8

I hereby certify that this correspondence is being deposited with the U.S. Postal Service as First Class Mail in an envelope addressed to: Mail Stop Amendment-Fee, Commissioner for Patents, P.O. Box 1450, Alexandria, Virginia 22313 on the date indicated below:

June 16, 2004
Date

Adienne White

Dear Commissioner:

I, Dr. Melvin C. Hunt, declare that:

1. I hold a degree of B.S. in Animal Husbandry from Kansas State University in Manhattan, Kansas that was obtained in 1965. I hold a degree of M.S. in Animal Science from Kansas State University that was obtained in 1970. I hold a degree of Ph.D. in Food Science from the University of Missouri in Columbia, Missouri that was obtained in 1973.

2. From 1973-1975, I worked as a research chemist for Tennessee Eastman Company in Kingsport, Tennessee in the health and nutrition division. Since 1975 to the present, I have held various professor positions at Kansas State University. Since 1991, I have been the Chair of

the Undergraduate Food Science Program at Kansas State University. I have taught several courses over the years at Kansas State University and some of those courses include the following: Meat Science, Processed Meat Operations, Advance Meat Science, Food Science Seminar, Topics in Meat Science and Muscle Biology, Meat Processing, and Livestock and Meat Evaluation. I have also performed numerous research projects in Meat Science and Muscle Biology including major emphasis on pigment chemistry, meat color, meat packaging, and factors effecting microbial soundness (shelf life) of meat. Thus, I have extensive experience in the processing of meat using modified atmosphere packaging.

3. My curriculum vita (attached as Exhibit A) details my professional affiliations related to animal science and meat science. I have served as President of the American Meat Science Association in 1995-1996, Chair of the Meat Science-Muscle Biology Section of National American Society of Animal Science ("ASAS"), Chair of the Midwestern ASAS Meat Science Section, and Chair of the Muscle Foods Division of the Institute of Food Technologists. I have been on the Editorial Board of the publication entitled "Journal of Muscle Foods." I also perform manuscript review for several peer-reviewed scientific publications including "Meat Science", "Journal of Muscle Foods", "Journal of Animal Science", and "Journal of Food Science

4. I assisted in preparing some of the information included in Pactiv's GRAS notice (Exhibit B) that was filed with the Food and Drug Administration (FDA) on August 29, 2001. The specific modified atmosphere packaging (MAP) system that was presented in the GRAS notice as a meat packaging system containing 0.4 vol.% CO and was referred to in the notice as Pactiv's ActiveTech® meat packaging system. The ActiveTech® meat packaging system placed meat in polystyrene trays, which were covered with oxygen-permeable, polyvinyl chloride (PVC)

overwraps. The wrapped trays of meat were then placed in an outer barrier bag. Air was removed and replaced with a blend of 0.4 vol.% CO, 30 vol.% carbon dioxide, and the balance being nitrogen.

5. I performed a series of tests on the effects of the ActiveTech® meat packaging system with CO on fresh meat color, color stability, and shelf life. The conclusions reached for the ActiveTech® meat packaging system with CO were: (a) the color of Pactiv's ActiveTech® meat packaging system using CO resulted in products that were equally red to products packaged with traditional oxygen permeable overwrap; (b) color deterioration of meat during simulated retail display in Pactiv's ActiveTech® meat packaging system using CO compared well to products packaged with traditional oxygen permeable overwrap; (c) bacterial growth was neither encouraged nor suppressed by adding CO to Pactiv's ActiveTech® meat packaging system; and (d) CO in the ActiveTech® meat packaging system neither masked spoilage, nor extended color life beyond the point of microbial soundness. I further concluded that Pactiv's ActiveTech® meat packaging system using 0.4 vol.% CO might be eligible for GRAS status.

6. The results of the testing were surprising to me because it was understood by those skilled in the art that CO fixes (creates a stable form of myoglobin that could mask spoilage) the color of the meat pigment to red. This is believed to be the reason on why CO had not been allowed to be used with fresh meat in the United States for many years. Pactiv's ActiveTech® meat packaging system using 0.4 vol.% CO, however, did not fix the color of the meat pigment to red. Rather, the meat pigment turned brown (discolored) in a pattern typical of retail meat in display but packaged in a standard supermarket format (foam tray and PVC overwrap). This was a novel result and was not at all obvious due to the current and long standing thought that meat

exposed to CO would develop a color that would mask spoilage. In other words, the pigment of the meat when exposed to CO would produce an extremely stable form of the pigment, but this did not happen in the Pactiv Active Tech® system.

7. Prior to Pactiv's ActiveTech® meat packaging system using 0.4 vol.% CO, there was a need in the industry to provide a solution that: (a) reduced the seasoning period (the critical time meat is exposed to low partial pressures of oxygen, which can seriously damage the pigment chemistry); (b) formed consistently a normal bloomed color with meats whose pigment is sensitive to metmyoglobin formation; and (c) avoided the fixing of too stable of a meat color, which can be unsafe and potentially dangerous, if the color stability was greater than the shelf life (microbial soundness) of the product. Such a solution was especially desirable for a centralized packaging facility where the meat would be shipped to distant locations. Pactiv's ActiveTech® meat packaging system using 0.4 vol.% CO was a new and novel approach that addressed these technological needs.

8. I hereby declare that all statements made of my own knowledge are true and that all statements made on information and belief are believed to be true; and, further, that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment or both under Section 1001 of Title 18 of the United States Code, and that such willful false statements may jeopardize the validity of the application or any patent issuing thereon.

Date: 6-14-04

Dr. Melvin C. Hunt
Dr. Melvin C. Hunt

MELVIN C. HUNT

Department of Animal Sciences and Industry
Weber Hall
Kansas State University
Manhattan, KS 66506-0201

Ph: 913-532-1232
Fax: 913-532-7059
Hhunt@oznet.ksu.edu

PERSONAL DATA:

Born: February 10, 1942

Married: Rae Jean Opie, August 20, 1965; Daughters: Paige and Holly

EDUCATION:

B.S. 1965 Animal Husbandry, Kansas State University, Manhattan, KS

M.S. 1970 Animal Science, Kansas State University, Manhattan, KS

Ph.D. 1973 Food Science, University of Missouri, Columbia, MO

PROFESSIONAL EXPERIENCE:

- 1991- Chair, Undergraduate Food Science Program
- 1984- Professor, Kansas State University: 50% Teaching - 50% Research
- 1978-84 Associate Professor, Kansas State University
- 1975-78 Assistant Professor, Kansas State University
- 1973-75 Research Chemist, Tennessee Eastman Company
- 1968-73 Grad Research Assistant, Kansas State and University of Missouri
- 1966-68 Taught high school chemistry and biology, Kinsley, KS

PROFESSIONAL AFFILIATIONS:

American Meat Science Association:

- President, 1995-96; Past-President, 1996-97
- Director and Executive Board, 1989-91
- Chair 1991 Reciprocal Meat Conference
- Parliamentarian
- Chair or member of numerous committees including:
 - Meat Color Guidelines, AMSA Teaching Award, Undergraduate Travel Award, Grad Student Poster Competition, Teaching Display, Resolutions, Meat Tenderness, Biochemistry-Biophysics, Packaging, Meat Color, Growth and Development, Reciprocation, Long Range Planning, Sustaining Membership, Endowment, and Research Priorities.

American Society of Animal Science:

- Chair and Chair-elect, Meat Science-Muscle Biology Section of National ASAS Meeting
- Chair, Midwestern ASAS Meat Science Section
- Editorial Board Journal Animal Science
- Teaching Award Committee, Midwestern ASAS Section

Institute of Food Technologists:

- Chair and Chair-elect of Muscle Foods Division, 1992-94
- Director of Muscle Foods Division
- Chair of Muscle Foods Nominating Committee
- Committee for two National Muscle Foods Symposia
- Journal of Food Science, Manuscript Review

CAST: Contributing member

Journal of Muscle Foods: Editorial Board

HONORARY AFFILIATIONS:

Phi Kappa Phi, Sigma Xi, Phi Tau Sigma, Gamma Sigma Delta, Alpha Zeta

HONORS:

- College of Agriculture Outstanding Faculty Award 1979
- College of Agriculture Outstanding Faculty Award 1982
- College of Agriculture Outstanding Faculty Award 1988
- College of Agriculture Outstanding Faculty Award 1998
- College of Agriculture Outstanding Academic Advisor 1983
- University Selection for Parents' Day Lecture 1979
- Outstanding Lecturer Award, ITAL, Campinas, Brazil 1981
- Honorary State Farmer Degree 1985
- Distinguished Teaching Award, Gamma Sigma Delta 1989
- Selected Instructor, National Food Science Satellite Program 1990
- Certificate of Meritorious Service, Kansas Ag Teachers Association 1992
- CASE Professor of the year, Kansas winner of national competition 1992
- Outstanding Advising Award, Gamma Sigma Delta 1994
- Distinguished Teaching Award, American Meat Science Association 1994
- Outstanding Food Scientist, Phi Tau Sigma 1996
- Outstanding KSU Instructor & Advisor Award, Mortar Board 1997
- Signal Service Award, American Meat Science Association 1997
- USDA Food & Agriculture Science Excellence in Teaching Award, 2000

DEPARTMENT, COLLEGE OF AG, AND UNIVERSITY ACTIVITIES:

- Faculty Advisor: Block and Bridle, 6 years
- Faculty Advisor: Food Science Club, 3 years
- Faculty Advisor: Animal Science Grad Student Association, 16 years
- Faculty Advisor: Ag Student Council, elected for 2 terms (4 years)
- Chair, Weber Hall Building/Renovation Project
- Chair, KSU Meat Science Faculty
- Coordinator of KSU Meat Research Labs
- ASI Graduate Student Selection Committee
- ASI Undergraduate Career Development Committee
- ASI Library Committee
- ASI Scholarship, Loans and Honors Committee
- Department Representative for Gamma Sigma Delta, 10 years
- Student Team Coordinator, ASI Quadrathlon Teams
- Agriculture Student of the Month Selection Committee
- Agriculture Faculty of the Semester Selection Committee
- College of Agriculture Course and Curriculum Committee, chair and member
- College of Agriculture Academic Standards Committee, chair and member
- College of Agriculture Commencement Committee
- University Faculty Senator, College of Agriculture, two terms (6 years)
- University Academic Affairs Committee
- University Coordinating Committee for United Way
- KAES NCR-121 Chair and Secretary: Food & Feed Safety in Animal Production
- Food Science Undergraduate and Graduate Steering Committees
- Chair, Non-Traditional Studies Advisory Committee
- Elected by peers to ASI Teaching Advisory Committee
- Chair, KSU Undergraduate Food Science Program: Coordinate all course & curriculum and policy matters, scholarship, internships, recruitment, and record keeping

INDUSTRY-EXTENSION ACTIVITIES:

- Numerous presentations at: MidWest Meat Processors Seminars, Kansas-Nebraska Curing and Sausage Short Courses, KSU Cattlemen's Day, KSU Swine Day
- Technical Assistance for: Tennessee Eastman Company, Ross Industries, Giant Food

Stores, Excel Corporation, IBP, Doskocil Companies, Tenneco Packaging, Farmland, National Beef, Cryovac, Buckhead Beef, Dupont, Kalsec, Wendy's, Greater Omaha Beef, Hormel

- State FFA Livestock Awards Selection Committee
- State FFA Star Farmer Selection Committee
- State FFA Public Speaking Contest Judge
- Kansas Jr. Livestock Carcass Contest Judge
- Kansas Meat Processor Cured Meat Show Judge
- Missouri Meat Processor Cured Meat Show Judge

TEACHING RESPONSIBILITIES:

Current Courses - KSU Campus:

- ASI 350 Meat Science. 3hr. Lecture-lab introductory meat science
Enrollment: Since 1979, 2031 students; currently running at maximum seating of 72
- ASI 610 Processed Meat Operations. 2hr. 50% responsibility, value-added processing
Enrollment: 6 to 12 undergraduate and graduate students; since 1988, 35 students
- ASI 930 Advanced Meat Science. 3hr. Team-taught, highest level meats course
Enrollment: Varies from 6 to 15 graduate students
- GENAG 500 Food Science Seminar. 1hr. Seminar for graduating seniors
Enrollment: Varies from 6 to 15 students

Current Courses - KSU Distance Learning Program:

- ASI 340 Principles of Meat Science. 2hr. Web-based course for Continuing Education
Enrollment: Since 1987, over 680 students
- GENAG 500 Food Science Seminar. 1hr. Seminar series for Distance Learning majors
Enrollment: 3 to 15 undergraduate students per year, Continuing Education
- GENAG 630 Food Science Problems. 1hr. Detailed written investigation of current topics
Enrollment: 2 to 8 students per year through Continuing Education

Previously Taught Courses:

- Topics in Meat Science and Muscle Biology
- Meats Judging Team (at University of Missouri)
- Meat Processing
- Livestock and Meat Evaluation
- Animal Agriculture and Consumers

INTERNATIONAL COURSE ACTIVITIES:

- Meat Science and Technology Short Course for Latin America, Institute for Food Technology, Campinas, Brazil, 6 weeks, one of two international lecturers
- Meat Science Facilities, University of Monterrey, Monterrey, Mexico
- Lecturer for five KSU International Meat Science Courses, International Meat and Livestock Program, Kansas State University
- Sabbatical leave, fall 1992, visiting scientist to Norwegian Food Research Institute, As
- Have attended 8 International Congresses of Meat Science and Technology

ADVISING RESPONSIBILITIES:

- Undergraduate Advisees: average of 26 for the last 10 years
- Graduate Students Supervised: Graduate Student Committees:
 - 12 Masters Students - 43 Masters
 - 6 PhD Students - 20 PhD
- Coordinate student-company relations for employment and internships for FSI

RESEARCH INTERESTS:

- Myoglobin chemistry and meat color, Methods of color measurement, Cooked meat color and food safety, Postmortem factors affecting meat quality, Collagen chemistry, Low-fat ground beef and processed meats; Six major company packaging projects funded since 1994 dealing with shelf life, color life, cold chain management, product palatability, and microbiology

PUBLIC AND COMMUNITY ACTIVITIES:

- Manhattan Optimist Club: committees for many youth activities
- Coach, Girls (16-18) ASA fast pitch softball traveling team
- Executive Committee, Riley County Extension Council
- Asst. Superintendent, sheep division, Riley County Fair
- Judge at Manhattan High School oratorical contest
- FarmHouse Fraternity, alumni board and committee work
- Snyder Award for Alumni Service, FarmHouse Fraternity
- Activities of First Presbyterian Church

Melvin C. Hunt
Professor
Department of Animal Sciences and Industry
Kansas State University

Refereed Journal Articles

- Hunt, M.C., R.A. Smith, D.H. Kropf and H.J. Tuma. 1975. Factors affecting showcase color stability of frozen lamb in transparent film. *J. Food Sci.* 40:637.
- Hunt, M.C. and H.B. Hedrick. 1977. Profile of fiber types and related properties of five bovine Muscles. *J. Food Sci.* 42:513.
- Hunt, M.C. and H.B. Hedrick. 1977. Histochemical and histological characteristics of bovine muscle from four quality groups. *J. Food Sci.* 42:578.
- Hunt, M.C. and H.B. Hedrick. 1977. Chemical, physical and sensory characteristics of bovine muscle from four quality groups. *J. Food Sci.* 42:716.
- Thomas, J.D., D.M. Allen, M.C. Hunt and C.L. Kastner. 1977. Nutritional regimen, post-slaughter conditioning temperature, and vacuum packaging effects on beef carcass and retail cut bacteriology. *J. Food Prot.* 40:678.
- Harrison, A.R., M.E. Smith, D.M. Allen, M.C. Hunt, C.L. Kastner and D.H. Kropf. 1978. Nutritional regimen effects on quality and yield characteristics of beef. *J. Anim. Sci.* 47:383.
- Loveday, H.D., M.E. Dikeman, M.C. Hunt and A.D. Dayton. 1978. Adipose tissue water related to bovine carcass composition. *J. Anim. Sci.* 47:606.
- Smith, M.E., C.L. Kastner, M.C. Hunt, D.H. Kropf and D.M. Allen. 1979. Elevated conditioning temperature effects on carcasses from four nutritional regimens. *J. Food Sci.* 44:158.
- Gutowski, G.H., M.C. Hunt, C.L. Kastner, D.H. Kropf and D.M. Allen. 1979. Vacuum aging, display, and level of nutrition effects on beef quality. *J. Food Sci.* 44:140.
- Fung, D.Y.C., C.L. Kastner, M.C. Hunt, M.E. Dikeman and D.H. Kropf. 1980. Mesophilic and psychrotrophic populations on hot-boned and conventionally processed beef. *J. Food Prot.* 43:547.
- Hayward, L.H., M.C. Hunt, C.L. Kastner and D.H. Kropf. 1980. Blade tenderization effects on beef longissimus sensory and Instron textural measurements. *J. Food Sci.* 45:925.
- Harrison, A.R., D.H. Kropf, D.M. Allen, M.C. Hunt and C.L. Kastner. 1980. Relationships of spectrophotometric reflectance measurements to beef muscle visual color. *J. Food Sci.* 45:1052.
- Burson, D.E., M.C. Hunt, D.M. Allen, C.L. Kastner and D.H. Kropf. 1980. Ration energy density and time on feed effects on beef longissimus palatability. *J. Anim. Sci.* 51:875.
- Fung, D.Y.C., C.L. Kastner, C-Y. Lee, M.C. Hunt, M.E. Dikeman and D.H. Kropf. 1981. Initial chilling rate effects on bacterial growth of hot-boned beef. *J. Food Prot.* 44:539.
- Wu, J.J., D.M. Allen, M.C. Hunt, C.L. Kastner and D.H. Kropf. 1981. Nutritional effects on beef collagen characteristics and palatability. *J. Anim. Sci.* 53:1256.
- Hall, J.B. and M.C. Hunt. 1982. Collagen solubility of A-maturity bovine longissimus muscle as affected by nutritional regimen. *J. Anim. Sci.* 55:321.
- Sleper, P.S., M.C. Hunt, D.H. Kropf, C.L. Kastner and M.E. Dikeman. 1983. Electrical stimulation effects on myoglobin properties of bovine longissimus muscle. *J. Food Sci.* 48:479.

- Axe, J.E. Bowles, C.L. Kastner, M.E. Dikeman, M.C. Hunt, D.H. Kropf and G.A. Milliken. 1983. Effects of beef carcass electrical stimulation, hot boning, and aging on unfrozen and frozen longissimus dorsi and semimembranosus steaks. *J. Food Sci.* 48:332.
- Lyon, M., C.L. Kastner, M.E. Dikeman, M.C. Hunt, D.H. Kropf and J.R. Schwenke. 1983. Effects of electrical stimulation, aging, and blade tenderization hot-boned beef psoas major and triceps brachii muscles. *J. Food Sci.* 48:131.
- Greathouse, J.R., M.C. Hunt, M.E. Dikeman, L.R. Corah, C.L. Kastner and D.H. Kropf. 1983. Ralgro implanted bulls: performance, carcass characteristics, longissimus palatability and carcass electrical stimulation. *J. Anim. Sci.* 57:355.
- Burson, D.B., M.C. Hunt, D.E. Schafer, D. Beckwith and J.R. Garrison. 1983. Effects of stunning method and time interval from stunning to exsanguination on blood splashing in pork. *J. Anim. Sci.* 57:918.
- Shivas, S.D., D.H. Kropf, M.C. Hunt, C.L. Kastner, J.L.A. Kendall and A.D. Dayton. 1984. Effects of ascorbic acid on the display life of ground beef. *J. Food Prot.* 47:11.
- Choi, Y.I., C.L. Kastner, M.E. Dikeman, M.C. Hunt and D.H. Kropf. 1984. Effects of electrical stimulation and hot boning on functional characteristics of preblended beef muscle in model systems. *J. Food Sci.* 49:867.
- Claus, J.R., D.H. Kropf, M.C. Hunt, C.L. Kastner and M.E. Dikeman. 1984. Effects of beef carcass electrical stimulation and hot boning on muscle display color of polyvinylchloride packaged steaks. *J. Food Sci.* 49:1021.
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- Shivas, S.D., C.L. Kastner, M.E. Dikeman, M.C. Hunt and D.H. Kropf. 1985. Effects of electrical stimulation, hot boning, and chilling on bull semimembranosus muscle. *J. Food Sci.* 50:36.
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- Flores, H.A., C.L. Kastner, D.H. Kropf and M.C. Hunt. 1986. Effects of blade tenderization and trimming of connective tissue on hot-boned, restructured, pre-cooked roast from cows. *J. Food Sci.* 51:1176.
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- Claus, J.R., M.C. Hunt and C.L. Kastner. 1989. Effects of substituting added water for fat on the textural, sensory, and processing characteristics of bologna. *J. Muscle Foods.* 1:1.
- Claus, J.R., M.C. Hunt, C.L. Kastner and D.H. Kropf. 1990. Low-fat, high-added water bologna: Effects of massaging, preblending, and time of addition of water and fat on the physical and sensory characteristics. *J. Food Sci.* 55:338.
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- Claus, J.R. and M.C. Hunt. 1991. Characteristics of low-fat, high-added water bologna formulated with textural modifying ingredients. *J. Food Sci.* 56:643.
- Whipple, G., M.C. Hunt, R.D. Klemm, D.H. Kropf, R.D. Goodband, J.L. Nelssen, R.H. Hines and B.R. Schrickler. 1992. Effects of porcine somatotropin and supplemental lysine on porcine muscle histochemistry. *J. Muscle Foods.* 3:217.
- Troutt, E.S., M.C. Hunt, D.E. Johnson, J.R. Claus, C.L. Kastner, D.H. Kropf and S. Stroda. 1992. Chemical, physical, and sensory characterization of ground beef containing 5 to 30 percent fat. *J. Food Sci.* 57:25.
- Troutt, E.S., M.C. Hunt, D.E. Johnson, C.L. Kastner and D.H. Kropf. 1992. Characteristics of low-fat ground beef containing texture-modifying ingredients. *J. Food Sci.* 57:19.
- Goll, S.J., C.L. Kastner, M.C. Hunt and D.H. Kropf. 1992. Effects of glucose and internal cooking temperature on the characteristics of low fat, pre- and post-rigor restructured beef roasts. *J. Food. Sci.* 57:834.
- Nold, R.A., J.A. Unruh, M.C. Hunt and C.W. Spaeth. 1992. Effects of implanting ram and wether lambs with zeranol at birth and weaning on palatability and muscle collagen characteristics. *J. Anim. Sci.* 70:2752.
- Warren, K.E., M.C. Hunt, C.L. Marksberry, O. Sörheim, D.H. Kropf and M.J. Windisch. 1992. Modified-atmosphere packaging with carbon dioxide for bone-in pork loins. *J. Muscle Foods.* 3:283.
- Nold, R.A., J.A. Unruh, C.W. Spaeth and M.C. Hunt. 1992. Effects of implanting ram and wether lambs with zeranol on pelt characteristics and removal. *Sheep Res. J.* 8:81.
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- Letelier, V., C.L. Kastner, P.B. Kenney, D.H. Kropf, M.C. Hunt, and C.M. Garcia Zepeda. 1995. Flaked sinew addition to low-fat cooked color. *J. Food Sci.* 60:1000.

Lavelle, C., M.C. Hunt, and D.H. Kropf. 1995. Display Life and Internal Cooked Color of Ground Beef from Vitamin E-Supplemented Steers. *J. Food Sci.* 60:1-4,6.

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Campbell, R.E. and M.C. Hunt. 1996. Yields, chemical composition, and value of beef shank tissues obtained using Baader processing. *J. Anim. Sci.* 74:786-789.

Warren, K.E., M.C. Hunt, D.H. Kropf, M.A. Hague, C.L. Waldner, S.L. Stroda, and C.L. Kastner. 1996. Chemical properties of ground beef patties exhibiting normal and premature brown internal cooked color. *J. Muscle Foods.* 7:303-314.

Warren, K.E., M.C. Hunt, and D.H. Kropf. 1996. Myoglobin oxidative state affects internal cooked color development of ground beef patties. *J. Food Sci.* 61:513-515,519.

Luchsinger, S.E., D.H. Kropf, C.M. Garcia Zepeda, M.C. Hunt, J.L. Marsden, E.J. Rubio Canas, C.L. Kastner, W.G. Kuecker, and T. Mata. 1996. Color and oxidative rancidity of gamma and electron beam-irradiated boneless pork chops. *J. Food Sci.* 61:1000-1005,1093.

Luchsinger, S.E., D.H. Kropf, C.M. Garcia Zepeda, E. Chambers IV, M.E. Hollingsworth, M.C. Hunt, J.L. Marsden, C.L. Kastner, and W.G. Kuecker. 1996. Sensory analysis and consumer acceptance of irradiated boneless pork chops. *J. Food Sci.* 61:1261-1266.

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Luchsinger, S.E., D.H. Kropf, E. Chambers IV, C.M. Garcia Zepeda, M.C. Hunt, S.L. Stroda, M.E. Hollingsworth, J.L. Marsden, and C.L. Kastner. 1997. Sensory analysis of irradiated ground beef patties and whole muscle beef. *J. Sensory Studies.* 12:105-126.

Garcia Zepeda, C.M., C.L. Kastner, J.R. Wolf, J.E. Boyer, D.H. Kropf, M. C. Hunt and C.S. Setser. 1997. Extrusion and low-dose irradiation effects on destruction of *clostridium sporogenes* spores in a beef-based product. *J. Food Prot.* 60:777-785.

Carmack, C.F., C.L. Kastner, M.C. Hunt, D.H. Kropf, C.M. Garcia Zepeda, and J.R. Schwenke. 1997. Sensory, chemical, and physical evaluation of reduced fat ground beef patties with "natural" flavorings. *J. Muscle Foods.* 8:199-212.

Luchsinger, S.E., D.H. Kropf, C.M. Garcia Zepeda, M.C. Hunt, S.L. Stroda, J.L. Marsden, and C.L. Kastner. 1997. Color and oxidative properties of irradiated whole muscle beef. *J. Muscle Foods.* 8:427-443.

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Hunt, M.C., O. Sørheim, and E. Slinde. 1999. Color and heat denaturation of myoglobin forms in ground beef. *J. Food Sci.* 64:847-851.

Powell, T.H., M.C. Hunt, and M.E. Dikeman. 1999. Enzymatic assay to determine collagen thermal denaturation and solubilization. *Meat Sci.* 54:307-311.

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Hunt, M.C. 1999. Distance learning methods. KSU College of Agriculture Focus Series.

Educational Materials

Course Syllabi:

Distance Learning - Complete course, *PRINCIPLES OF MEAT SCIENCE* in "any-time or real-time" distance learning format via audiotapes, teleconferencing, on-line discussions involving collaborative learning, problem solving and critical thinking via the Internet.

In-Class - Laboratory Materials for *MEAT SCIENCE*, a series of 14 study guides for lab exercises
- Laboratory Materials for *PROCESSED MEAT OPERATIONS*

Other Materials and Activities:

USDA Grant: - Expanding Undergraduate Education for Food Industry Personnel via Technology.
1994-96 USDA Challenge Grant Program, \$79,479

Web-based Course - Principles of Meat Science, KSU Division of Continuing Education

Color Guides - Ground Beef Patty Cooked Color Guide
- Cured Meat Color Guide
- Cooked Pork Chop Color Guide
- Ground Pork Patty Cooked Color Guide

Science Series - Lesson Plans for: Promoting Ag Science for Secondary Schools
Developing New Meat Products
Color Chemistry in Meat Products
Meat Packaging Exercises for High School Students

Slides Series: - Unraveling the Mystery of Premature Browning in Cooked Ground Beef Patties
- Doneness of Cooked Ground Beef
- Dynamics of Conversion of Myoglobin Forms
- Role of Pigment Layers in Influencing Surface Meat Color
- Spray Chilling of Carcasses
- Don't be Broken-Hearted because of High-fat in Ground Beef
- Commercial Sausage, Ham and Bacon Production
- Food Science at KSU
- ASI Quadrathlon - why I should participate
- Updated: Muscle-Bone Anatomy; Beef-Pork-Lamb Cut Identification

Video Tapes: - Beef Carcass Electrical Stimulation and Hot Boning
(Edited with M. E. Dikeman)

Store Survey: - Out-of-class assignment for Analysis of Retail Meat Section of Grocery Stores

Diet Survey: - Out-of-class assignment for computerized class project of Nutritional Value of Muscle Foods in the student's diet

Current topic: Survey - Out-of-class assignment for critically analyzing printed literature on a variety of livestock and meat industry topics

Web Sites: - Out-of-class assignment for evaluation and collection of scientific facts about muscle biology and meat science

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- Troutt, S.E. 1990. A chemical, physical and sensory characterization of low-fat ground beef. M.S. Thesis, Kansas State University.
- Warren, K.E. 1990. Modified atmosphere packaging with 100% carbon dioxide for bone-in pork loins. M.S. Thesis, Kansas State University.
- Hague, M.A. 1992. Internal and expressible juice color of cooked ground beef patties. M.S. Thesis, Kansas State University.
- Londono Villegas, J.F. 1993. Effects of realimentation and trenbolone acetate implantation of cull cows on tenderness and cooked color. M.S. Thesis, Kansas State University.
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- Wendelburg, J. 2001. Physical, chemical and microbial qualities of blade tenderized prime rib. MS Thesis.
- Ballard, C. 2002. Carbon monoxide in modified atmosphere packaging. MS Thesis.

ERIC F. GREENBERG
ATTORNEY AT LAW

ERIC F. GREENBERG
Of Counsel
Ungaretti & Harris

Contains Confidential Business Information

August 29, 2001

Division of GRAS Notice Review
Office of Food Additive Safety
Center for Food Safety and Applied Nutrition
Food and Drug Administration
200 C St., SW
Washington, DC 20204

Re: NOTIFICATION OF CLAIM FOR GENERAL RECOGNITION OF
SAFETY OF CARBON MONOXIDE IN A MODIFIED ATMOSPHERE
SYSTEM FOR PACKAGING FRESH MEAT, submitted by Pactiv
Corporation


To the FDA:

Enclosed is the NOTIFICATION OF CLAIM FOR GENERAL RECOGNITION OF
SAFETY OF CARBON MONOXIDE IN A MODIFIED ATMOSPHERE SYSTEM FOR
PACKAGING FRESH MEAT, submitted by Pactiv Corporation, 1900 West Field Court,
Lake Forest, Illinois 60045, c/o the undersigned counsel, consisting of pages 000001.001
through 000250.

Please note that this submission contains Confidential Business Information that
Pactiv Corporation desires not to be revealed to Freedom of Information Act
requestors and other members of the public. In the first copy of the submission
following this letter, the Confidential Business Information has been redacted. For
ease of reference, a list indicating which pages contain redactions is attached.

Five complete copies of the submission are enclosed, including the one that has been
redacted.

If you have any questions, please contact me at 312 977-4647.

Sincerely,

Eric F. Greenberg
Enc.

000001.001

REDACTIONS

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Notification of Claim for General Recognition of Safety of Carbon Monoxide in A
Modified Atmosphere System for Packaging Fresh Meat

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Attachment 4	"Evaluation of Beef Steaks and Ground Beef in the Pactiv ActiveTech Packaging System: Effects of Carbon Monoxide in the Package Atmosphere", Hachmeister, K; Hunt, M.; Milliken, G; May 2001	000157
Attachment 5	Certificate of Conformance, Carbon monoxide, Haun Welding Supply, Inc., Syracuse, New York, May 8, 2001.	000190
Attachment 6	"Standard Practice for Analysis of Reformed Gas by Gas Chromatography", ASTM D 1946-90 (Reapproved 2000)	000192
Attachment 7	Daren Cornforth, Ph.D., letter and curriculum vitae	000198
Attachment 8	Vasilios H. Frankos, Ph.D., letter and curriculum vitae	000205
Attachment 9	Melvin C. Hunt, Ph.D., letter and curriculum vitae	000221
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- Attachment 10 Oddvin Sørheim, Ph.D., letter and curriculum vitae

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List of Attachments

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ATTACHMENT 1



PACTIV
Advanced Packaging Solutions

August 9, 2001

Division of GRAS Notice Review
Office of Food Additive Safety
Center for Food Safety and Applied Nutrition
Food and Drug Administration
200 C St, SW
Washington, DC 20204

Pactiv Corporation
Technology Center
2651 Brickyard Road
Canandaigua, New York 14424-1026

**Re: Authorization of counsel regarding
NOTIFICATION OF CLAIM FOR GENERAL
RECOGNITION OF SAFETY OF CARBON MONOXIDE
IN A MODIFIED ATMOSPHERE
SYSTEM FOR PACKAGING FRESH MEAT**

To the FDA:

Please take note that Pactiv Corporation, with headquarters at 1900 West Field Court, Lake Forest, Illinois, 60045, authorizes its attorney, Eric F. Greenberg, 3500 Three First National Plaza, Chicago, Illinois 60602, to represent it and communicate on its behalf in all matters regarding Pactiv's NOTIFICATION OF CLAIM FOR GENERAL RECOGNITION OF SAFETY OF CARBON MONOXIDE IN A MODIFIED ATMOSPHERE SYSTEM FOR PACKAGING FRESH MEAT.

Sincerely,

For PACTIV CORPORATION

By: Vinod K. Luthra
General Manager
New Business Development
2651 Brickyard Road
Canandaigua, New York 14424

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Summary regarding Pactiv Corporation

Pactiv Corporation, 1900 West Field Court, Lake Forest, Illinois, is a leading provider of advanced packaging solutions to customers around the world. The company employs 17,000 people in 87 facilities worldwide. Annual revenues exceed \$3 billion.

Pactiv manufactures, markets and sells plastic and paper-based consumer products and food/foodservice packaging as well as protective and flexible packaging. Approximately 80% of its revenue comes from products made from different types of plastics, with the balance from paper and aluminum products.

The company's products include a wide range of items for consumers, food processors, supermarkets, foodservice entities, and the construction, automotive, computer, electronic, furniture and durable goods industries. The consumer products are sold under such recognized brand names as Hefty®, Baggies®, Hefty One-Zip®, Kordite™ and E-Z Foil®.

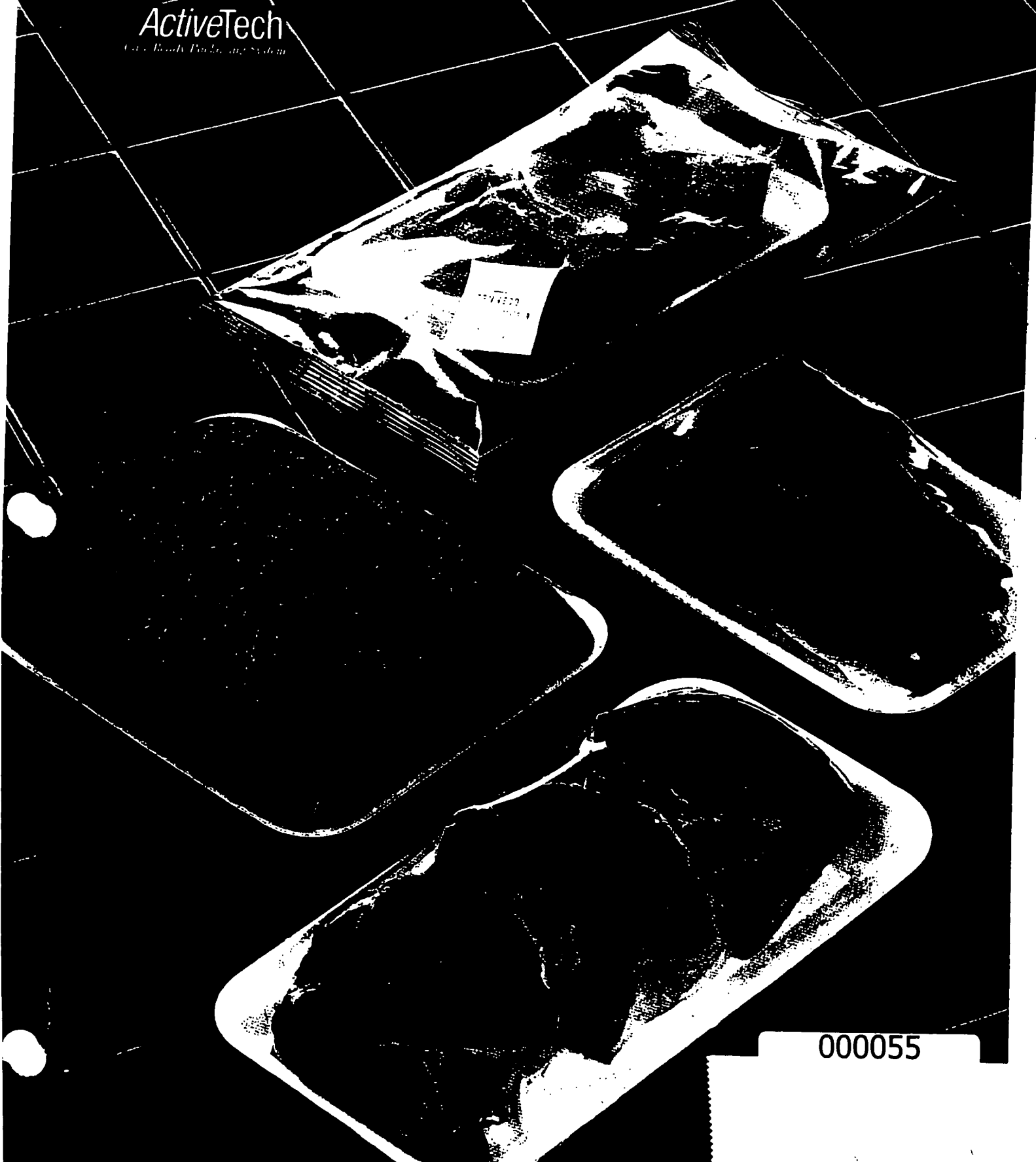
Pactiv further fuels internal growth by developing and commercializing proprietary new products and by designing value-added product-line extensions. In 1998, the consumer products and food/foodservice packaging business introduced over 80 new products and product-line extensions. In the protective and flexible packaging business, where custom design services drive revenues, Pactiv developed over 500 custom product applications in 1998. New product innovations include ActiveTech™ packaging, a proprietary modified atmospheric package used by food processors for case-ready meat.

ATTACHMENT 2

ActiveTechTM *Case Ready Packaging System*

For Case Ready Applications

ActiveTechTM
Case Ready Packaging System

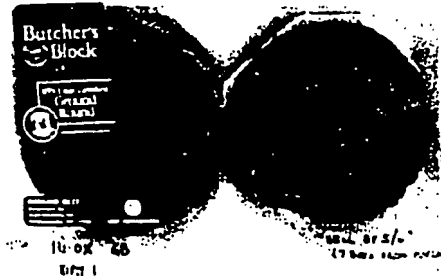


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First Display Day

Hi-Oxygen

20 Days Stored

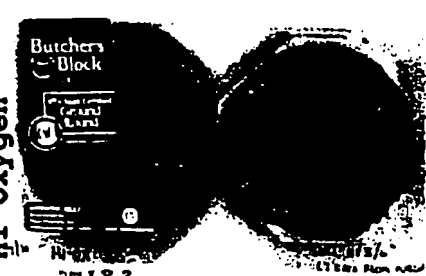


Beef: 80% Lean Ground Beef
Hi-OX, 20 Days Stored in "Short Gas", and Fresh
(0.4% CO/ 35% CO2/ 64.6% N2)

Third Display Day

Hi-Oxygen

20 Days Stored



Beef: 80% Lean Ground Beef
Hi-Ox, 20 Days Stored in "Short Gas", and Fresh
(0.4% CO/ 35% CO2/ 64.6% N2)

Second Display Day

Hi-Oxygen

20 Days Stored

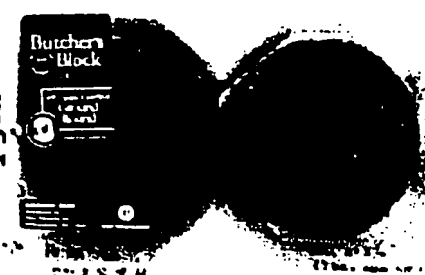


Beef: 80% Lean Ground Beef
Hi-Ox, 20 Days Stored in "Short Gas", and Fresh
(0.4% CO/ 35% CO2/ 64.6% N2)

Fourth Display Day

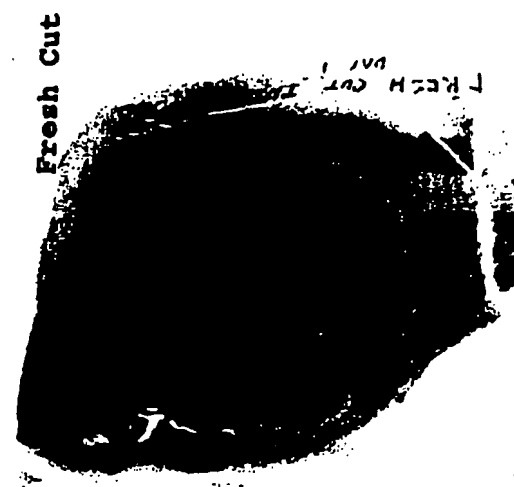
Hi-Oxygen

20 Days Stored



Beef: 80% Lean Ground Beef
Hi-Ox, 20 Days Stored in "Short Gas", and Fresh
(0.4% CO/ 35% CO2/ 64.6% N2)

First Display Day



Fresh Cut

Second Display Day



Fresh Cut



20 Days Stored

Beef: Top Round Steak
Fresh Cut and 20 Days Stored in "Short Gas"
(0.4% CO/ 35% CO2/ 64.6% N2)

Third Display Day



20 Days Stored

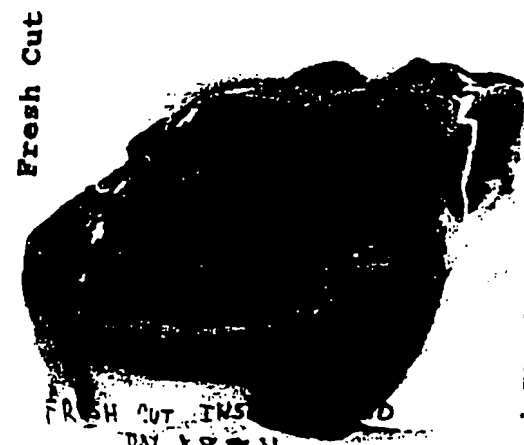


Fresh Cut

Fourth Display Day



20 Days Stored



Fresh Cut

Beef: Top Round Steak
Fresh Cut and 20 Days Stored in "Short Gas"
(0.4% CO/ 35% CO2/ 64.6% N2)

Beef: Top Round Steak
Fresh Cut and 20 Days Stored in "Short Gas"
(0.4% CO/ 35% CO2/ 64.6% N2)

The pages immediately following illustrate:

1. On the top of the photograph on the first page, an example is shown of the structure utilized for both ActiveTech™ and AT2001 incorporating tray, flexible overwrap, outer bag and activated oxygen scavenging sachet.
2. The second page of photographs show examples of ground meat color during the first, second, third and fourth days of display after packaging in (1) Hi-oxygen; (2) AT2001 atmosphere (referred to in captions as "Short Gas", 0.4% CO/35% CO₂/64.6 N₂), and after being held in that atmosphere for 20 days; and (3) Fresh.
3. The third page of photographs show examples of whole muscle meat (top round steak) color during the first, second, third and fourth days of display after packaging in (1) AT2001 atmosphere (referred to in captions as "Short Gas", 0.4% CO/35% CO₂/64.6 N₂), and after being held in that atmosphere for 20 days; and (2) Fresh cut.

ATTACHMENT 3

**APPLICATION FOR ASSESSMENT OF THE FOOD ADDITIVE CARBON MONOXIDE
(CO) PRIOR TO ITS AUTHORIZATION**

(This application is based on the document "Presentation of an application for assessment of a food additive prior to its authorisation", Office for Official Publications of the European Communities, Luxembourg, 1989, ISBN 92-826-0135-8).

PART I. ADMINISTRATIVE DATA

I.1. Applicants: Altogether, the two applicants represent the total meat industry in Norway

Applicant no. 1:

The name of the applicant:

Norsk Kjøttssamvirke (Norwegian Meat Cooperative)

Address:

Lorenveien 37
P.O.Box 360 Økern
0513 Oslo, Norway

Other means of communication:

Telephone: +47 22 09 21 00
Fax: +47 22 15 59 08

Applicant no. 2:

The name of the applicant:

Kjøttbransjens Landsforbund (The Norwegian Independent Meat Association) – represents the private meat industry in Norway

Address:

Karoline Kristiansensvei 2, Fyrstikktorget
P.O.Box 6279 Etterstad
0603 Oslo, Norway

Other means of communication:

Telephone: +47 23 24 44 70, Fax: +47 23 24 44 80

1.2. The name of the manufacturer(s) of the substance:

RIVOIRA S.P.A., Stabilimento Chivasso gas, Via cardinal Massaia 75L, I-10147 Torino, Italy

1.3. The name of the person responsible of the dossier:

Research director Truls Nesbakken, Norwegian Meat Research Centre, P. O. Box 396 Økern, 0513 Oslo, Norway. Telephone +47 22 09 23 99, Mobile phone +47 91 87 81 46, Fax +47 22 22 00 16, e-mail: truls.nesbakken@fagkjott.no

1.4. The table of contents of dossier

This dossier is sent through the Norwegian Food Control Authority (Statens næringsmiddeltilsyn). Together with this document follow as enclosures:

- 1) Nissen, H., Alvseike, O., Bredholt, S., Holck, A. and Nesbakken, T. (submitted) Packaging of ground beef in an atmosphere with high carbon dioxide and low carbon monoxide restrains growth of *Yersinia enterocolitica*, *Listeria monocytogenes* and *Escherichia coli* O157:H7. Int. J. Food Microbiol. As long as this work is not published, please handle this information with care.
- 2) Nissen, H., Alvseike, O., Bredholt, S., Holck, A. and Nesbakken, T. (1999) Packaging of ground beef in an atmosphere with low carbon monoxide and high carbon dioxide restrains growth of *Escherichia coli* O157:H7, *Listeria monocytogenes*, *Yersinia enterocolitica* and *Salmonella diarizonae*. In: Tuijelaars, A.C.J., Samson, R.A., Rombouts, F.M., Notermans, S., (Eds.), Food Microbiology and food safety into the next millenium. Proceedings of the Seventeenth International Conference of the International Committee on Food Microbiology and Hygiene (ICFMH), 13-17 September 1999, Veldhoven, The Netherlands, pp. 285-286.
- 3) Solheim, R. (1996) Consumer purchase probability of beef and pork packaged in different atmospheres. Report, Matforsk, 10 pp.
- 4) Sørheim, O. (1996) Discoloration of meat as an indicator of leakages in packages containing a CO gas mixture. Report, Matforsk, 5 pp.
- 5) Sørheim, O., Aune, T. and Nesbakken, T. (1997a) Technological, hygienic and toxicological aspects of carbon monoxide used in modified-atmosphere packaging of meat. Trends in Food Sci. Technol. 8, 307 - 312
- 6) Sørheim, O., Nissen, H., and Nesbakken, T. (1999) The storage life of beef and pork packaged in an atmosphere with low carbon monoxide and high carbon dioxide. Meat Sci. 52, 157 - 164

- 7) Letter from the director of Swedish Meats (which is the organisation of the Swedish meat cooperative) supporting the Norwegian meat industry's application to the EU Commission
- 8) Letter from the director of Swedish Meat Trade Association (which is the organisation of the private meat industry in Sweden) supporting the Norwegian meat industry's application to the EU Commission
- 9) Letter from the director of the Danish Pig Producers and Slaughterhouses, Copenhagen, Denmark supporting the Norwegian meat industry's application to the EU Commission
- 10) Letter from the Spanish Meat Industry's Association supporting the Norwegian meat industry's application to the EU Commission
- 11) Letter from the Finnish Meat Research Institute supporting the Norwegian meat industry's application to the EU Commission

PART II. TECHNICAL DATA

II.1. Name of the substance

- names in the IUPAC nomenclature: carbon monoxide
- other names (usual name/trade name/synonyms: carbon oxide, carbon monoxide)
- abbreviations: CO
- CAS number (if this has been attributed): 630 - 08 - 0

II.2. Specification of the substance

- composition (% , m/v, mg/kg), e.g. in the case of heterogeneous products): 100%
- empirical and structural formula: CO
- molecular weight: 28.010 g/mole
- degree of purity (%): higher than 99.3%
- nature of known impurities/percentage of significant and main impurities: (< 0.7%):

Impurities:	Concentration:
Oxygen + Argon	< 2500 vpm
Water	< 20 vpm
THC (Total hydrocarbons)	< 500 vpm
Hydrogen + Nitrogen	< 3500 vpm
Carbon dioxide	< 500 vpm

- physical form (liquid, powder, etc.): gas

- solubility (e.g. aqueous, organic solvents, lipid):

solubility in water, 0°C, a CO partial pressure of 101.325 kPa = 3.537 cm³/100 cm³. Solubility in organic solvents and lipid: not relevant – see Part II.6 Exposure

- other data that the applicant believes may be useful to identify the substance (e.g. physico-chemical properties, analytical data on differences between batches):

Thermodynamic properties of carbon monoxide as ideal gas at 25°C:

Heat capacity, c_p : 29.142 J/(mol * °K)

Entropy, S: 197.543 J/(mol * °K)

Enthalpy: 8.669 kJ/mol

- information on the microbiological characteristics, in particular on the possible presence of pathogens and bacterial or mycotoxins: not relevant

II.3. Manufacturing process

- Information on the method of manufacture (i.e. the process by which the raw materials are converted to the finished product):

The CO-gas is bought from RIVOIRA S.P.A., Torino, Italy (see I.2.). Hydro Rjukan Næringspark; P.O.Box 43-44, N-3661 Rjukan, Norway, makes the two CO-gas mixtures which are used in the Norwegian meat industry. They are called "Pakkemix NC1" and "DNC 29.7 – 0.3"

1) Pakkemix NC1 = 1.0% CO + 99% N₂

The production:

- evacuation of an empty cylinder to under 10 mbar
- flushing with N₂, quality 5.0
- repeat the evacuation of an empty cylinder to under 1 mbar
- manometric filling with CO, quality 2.3
- manometric filling with N₂ quality 5.0
- every tenth cylinder is analysed with gas chromatograph (GC) and thermoconductivity detector (TCD)

The pressure of the cylinder is 200 bar.

2) $\text{DNC } 29.7 - 0.3 = 0.3\% \text{ CO} + 29.7\% \text{ N}_2 + 70\% \text{ CO}_2$

The production:

- a) evacuation of an empty cylinder to under 10 mbar
- b) flushing with N_2 , quality 5.0
- c) repeat the evacuation of an empty cylinder to under 1 mbar
- d) manometric filling with CO , quality 2.3
- e) manometric filling with N_2 , quality 5.0
- f) manometric filling with CO_2 , quality 3.0
- g) every tenth cylinder is analysed with gas chromatograph (GC) and thermoconductivity detector (TCD)

The pressure of the cylinder is 50 bar..

II.4. Methods of analysis

- analytical methods to describe the substance, evaluate its purity and measure its physico-chemical and microbiological characteristics:

Oxygen + Argon, and Hydrogen + Nitrogen: Gas chromatograph with thermoconductivity detector (TCD) – min. detect. limits 10 vpm (RIVOIRA S.P.A., Stabilimento Chivasso gas, Via cardinal Massaia 75L, I-10147 Torino, Italy)

THC: Gas chromatograph with flame ionization detector (FID) – min. detect. limit 0.5 vpm (RIVOIRA S.P.A., Stabilimento Chivasso gas, Via cardinal Massaia 75L, I-10147 Torino, Italy)

Carbon dioxide: Gas chromatograph with helium ionization detector (HID) – min. detect. limit 0.5 vpm (RIVOIRA S.P.A., Stabilimento Chivasso gas, Via cardinal Massaia 75L, I-10147 Torino, Italy)

Water: Specific water analyzer – min. detect. limit 0.1 vpm (RIVOIRA S.P.A., Stabilimento Chivasso gas, Via cardinal Massaia 75L, I-10147 Torino, Italy)

- analytical methods for the determination of the additive and its degradation products (where relevant), in the foodstuff of which the substance is to form part:

The isotope, C^{14} , might be used for measuring CO before and after heat treatment (Watts et al., 1978). Spectrophotometry is used to measure carboxymyoglobin at 540 and 577 nm (native) or 425, 542 and 570 nm (denaturated) (El-Badawi et al., 1964; Comforth, 1994).

II.5. Justification for the additive

- intended use and purpose:

Fresh red meat (mainly beef, pork and lamb, but also horse, goat, reindeer, game etc.) packaged in an atmosphere with 60 - 70% carbon dioxide (CO₂), 30 - 40% nitroge..) and < 0.5% carbon monoxide (CO) (high CO₂/low CO mixture).

Gas mixtures with low concentrations of CO and high concentrations of CO₂ provide a combination of a long microbiological shelf life and a stable bright red colour of meat (Sorheim et al., 1999).

- the quantity to be added to specific foods and the residues in food: < 0.5% CO.
- investigations on the efficacy of the substance for the intended effect at the level proposed:

The main function of low levels of CO in modified atmospheres (MAs) is to give a stable, cherry red colour of the meat through strong binding of CO to myoglobin and formation of carboxymyoglobin (El-Badawi, 1964). Although a substantial increase in the shelf life of meat can be obtained by using various MAs, it is often limited by discolouration due to oxidation of myoglobin to metmyoglobin. This discolouration can be prevented by including a small fraction of CO in the gas mixture. Carboxymyoglobin is more resistant to oxidation than oxymyoglobin, due to the stronger binding of CO to the iron-porphyrin site on the myoglobin molecule (Wolfe, 1980). CO in concentrations of 1 - 5% had the ability to increase metmyoglobin reduction, even in the presence of air (Lanier et al., 1978).

The high CO₂/low CO mixture and absence of O₂ provides a unique combination of a long microbiological shelf life (caused by the high CO₂ level) and a stable cherry red colour (caused by the low CO level). CO₂ inhibits growth of many microorganisms, but it has no effect *per se* on the colour of the meat (Renerre and Labadie, 1993). This gas is absorbed in meat and fat tissue at a ratio of approximately 1 litre gas per kg tissue (Gill, 1988). N₂ affects neither the microbiology nor the colour of the meat, but prevents collapse of the packages because it is not absorbed in the product. O₂ supports the growth of aerobic microorganisms, and removal of O₂ from the MA will therefore extend the microbiological shelf life. The shelf life of meat is considerably longer in the high CO₂/low CO mixture than in the commonly used atmosphere of high oxygen (O₂) with approximately 70% O₂ and 30% CO₂. Consumption of meat treated with the high CO₂/low CO mixture will result only in negligible levels of carboxyhaemoglobin in blood. It is highly improbable that CO from packaging of meat will present a toxic threat to the consumer (Sorheim et al., 1997a).

Shelf life in the high CO₂/low CO mixture in comparison with alternative packaging methods:

Ground beef, beef loin steaks and pork chops were packaged in MAs of 0.4% CO/ 60% CO₂/ 40% N₂ (high CO₂/low CO mixture) and 70% O₂/ 30% CO₂ (high O₂ mixture). In addition ground beef was packaged in clipped chub packs, beef loin steaks were vacuum packaged, and pork chops were packaged in an atmosphere of 60% CO₂/ 40% N₂ with each pack containing an

O₂ absorber. The packs were stored in the dark at 4°C or 8°C for up to 21 days. Meat in the high CO₂/low CO mixture had a stable bright red colour. The storage lives in this gas mixture at 4°C, as limited by off-odours, were 11, 14 and 21 days for ground beef, beef loin steaks and pork chops, respectively. The high O₂ mixture resulted in an initially bright red to red colour of the meat, but the colour was unstable and off-odours developed rapidly. The off-odours probably were caused by *Brochothrix thermosphacta*, which grew in all meat types, and in ground beef by pseudomonads also. Meat stored in chub packs, vacuum packs and 60% CO₂/40% N₂ with an O₂ absorber developed off-odours and microflora similar to those of meat in low CO/high CO₂ mixture with however less acceptable colours or appearances. These results show that a low CO/high CO₂ atmosphere is effective for preserving retail-ready meat (Sørheim et al., 1999).

Aspects of spoilage:

Consumers may evaluate the shelf life of packaged meat based on its colour. A possible negative aspect of using CO in modified atmosphere packaging (MAP) of retail meat is a concern that the consumer might misjudge the product, because the microbiological status may be masked by the stable cherry red carboxymyoglobin colour (Knopf, 1980). However, the consumer is able to detect spoilage by off-odour (Sørheim et al., 1999). This is in contrast to ready to eat products, such as cooked, sliced vacuum or gas packaged meat, gas packaged vegetables and vacuum-packaged cheeses where the consumers often have to taste it before judging the product as unacceptable. As ready to eat products, they also represent a higher risk than fresh meat packed in the high CO₂/low CO mixture which is heat treated before consumption. In the current low concentrations, below 0.5%, CO *per se* seems to have no or only minor effects on bacteria and the shelf life of the meat. The combination of CO with high concentrations of CO₂, for example 60 - 70%, is necessary for microbiological control. MAP enables centralised packaging operations with quality control, but MAP alone is no guarantee for the shelf life of the product. Sufficient shelf life can only be obtained through a proper quality control of raw materials, production, packaging, chill chain and retail conditions (Sørheim et al., 1999).

Pathogens in the high CO₂/low CO mixture in comparison with alternative packaging methods:

Growth of the pathogens *Yersinia enterocolitica*, *Listeria monocytogenes*, *Escherichia coli* O157:H7 and strains of *Salmonella* was compared in ground beef packed in high CO₂/low CO mixture, high O₂ mixture and in chub packs. The ground beef was inoculated with rifampicin or nalidixic acid/streptomycin-resistant strains (final concentration 10²-10³ bacteria/g) and stored at 4°C and 10°C for up to 14 days. At 4°C the shelf life based on stable colour and reduced background flora was prolonged for the high CO₂/low CO mixture, compared to the two other packaging methods, but at 10°C the shelf life was < 8 days for all the packaging methods. Growth of *Y. enterocolitica* was nearly totally inhibited both at 4°C and 10°C in the high CO₂/low CO mixture, while the bacterial numbers in the samples packed in the high O₂ mixture increased from about 5x10² bacteria/g at day 0 to about 10⁴ at day 5 at 4°C and to 10⁵ at 10°C. Growth in the chub packs was even higher. *L. monocytogenes* showed very little growth at 4°C in all treatments. At 10°C there was slow growth of *L. monocytogenes* from about 5x10³ bacteria/g to about 10⁴ at day 5 in the high CO₂/low CO mixture, while the numbers in the high O₂ mixture and the chub packs were about 10 times higher. Growth of *E. coli* O157:H7 at 10°C in the ground beef was nearly totally inhibited in both the high CO₂/low CO mixture and the high O₂ mixture. Growth of *E. coli* O157:H7 in the chub packs was higher reaching 10⁵ bacteria/g on day

5. The *Salmonella* strains (*S. Typhimurium*, *S. Dublin*, *S. Enteritidis* and *S. enterica* «61:k1, 5.(7)») in the ground meat stored at 10°C for 5 and 7 days, grew to a higher number in the high CO₂/low CO mixture than in the high O₂ mixture. The present study shows that the prolonged shelf life at 4°C did not increase growth of *Y. enterocolitica* and *L. monocytogenes* in ground beef stored in the high CO₂/low CO mixture, but the observed growth of strains of *Salmonella* in this mixture and in chub packs at the abuse temperature of 10°C does emphasise the importance of temperature control during storage (Nissen et al. 1999; Nissen et al., submitted).

Consumer purchase probability:

Ground beef, beef loin steaks and pork chops were packaged in high CO₂/low CO mixture and high O₂ mixture. In addition ground beef was packaged in clipped chub packs, beef loin steaks were vacuum packaged, and pork chops were packaged in an atmosphere of 60% CO₂/ 40% N₂ with each pack containing an O₂ absorber. The purchase probability data were collected by interviewing 126 consumers usually purchasing meat and meat products. The consumers visually compared the samples within each type of meat after 4 days storage at 4°C. The consumers preferred ground beef packaged in the high CO₂/low CO mixture or the high O₂ mixture compared to ground beef packaged in clipped chub packs. Purchase probability increased when pork chops were packaged in the high CO₂/low CO mixture. Pork chops in packs containing an O₂ absorber, were rated lowest in purchase probabilities. The purchase probability for beef loin steaks was similar when packaged in the high CO₂/low CO mixture or the high O₂ mixture, and these products were preferred compared to beef loin steaks packaged in vacuum (Solheim, 1996)

- documentation on the need for the additive:

Alternatives to the high CO₂/low CO mixture:

The most common MA for retail packaging of meat today contains O₂ at high concentrations combined with CO₂, approximately 70% O₂/ 30% CO₂. The shelf life of meat in high O₂ atmospheres in commercial practice, typically at temperatures of 6 - 8°C, is about 7 days, limited by both microbiological spoilage and discolouration. Meat stored in the high O₂ mixture is often spoiled by bacteria like *Brochothrix thermosphacta* and pseudomonads (Gill, 1996). In MAs with high concentrations of O₂, the meat normally maintains its bright red oxymyoglobin colour for 4 - 7 days before the colour starts deteriorating into grey/brown due to formation of metmyoglobin (Sørheim et al., 1999). This length of time is often not considered sufficient for displaying and selling the product to 4.5 mill. inhabitants all along the distance from Kristiansand in the south to Kirkenes in the north of Norway (about 2700 km) corresponding to the distance from Oslo to Rome (about 2600 km)!

Using high CO₂, MAs of either CO₂ alone or CO₂/N₂ with up to 70% CO₂ increases the microbiological shelf life of the meat compared to MAs of high O₂. The absence of O₂ combined with the presence of CO₂ retard the microbiological growth. Unfortunately, the colour of the meat in MAs of CO₂ is less satisfactory, either as purple deoxymyoglobin or as grey/brown metmyoglobin. The meat inevitably discolours when concentrations of O₂ are low. Tolerance levels for avoiding metmyoglobin formation are less than 0.1% O₂ for beef (Gill and McGinnis, 1995) and 0.5% O₂ for pork (Sørheim et al., 1997b). These low O₂ levels, particularly for beef, are difficult to achieve in most commercial packaging operations, because small fractions of air will be

incorporated in the MAs of the packages. MAs with high CO₂ concentrations seem to be useful for retail packaging when combined with low concentrations of CO for stabilisation of myoglobin and the meat colour.

Vacuum is a packaging method commonly used for bulk storage, transportation and export of meat. However, vacuum has not been a successful method for retail packaging of meat, because of the purple deoxymyoglobin colour of the meat and the visible exudate that occurs in the packages (Bruce, 1990; Gill, 1996). Meat packaged in vacuum can not be presented in the bright red oxymyoglobin state, which depends on the presence of high concentrations of O₂ (Gill, 1996; Taylor et al., 1990), or alternatively as cherry red carboxymyoglobin with CO included in the MA.

Hazard for workers:

One of the objections raised against CO as a component of a packaging gas is the potential hazard it might represent for the workers in the meat plants. Using pure CO for mixing in the plant would certainly be such a risk, however, CO is delivered as a premixture (DNC 29.7 - 0.3) or in a 1% mixture together with 99% N₂ (Pakkemix NC1), which is the practice of gas suppliers to the Norwegian meat industry. This way of supplying CO is recognised to be a very safe handling procedure by the health authorities. MAs with concentrations of 60 - 70% O₂ must be handled carefully, because they are explosive gas mixtures. Strict safety regulations apply to explosive gas mixtures, resulting in additional costs of equipment and packaging operations. The benefit of the CO mixture is that it carries no risk or handling costs due to risk of explosion.

Experience of the Norwegian meat industry:

Despite the long term knowledge of CO and its many positive properties as a component of MAs for meat, the CO mixture has not been adopted to any large extent by the global meat industry. In many countries, like the US and EU, CO is presently not permitted for use in MAP of meat (Cornforth, 1994; European Parliament and Council Directive, 1995). However, the Norwegian food control authority has derogated from the EU directive for a two years period. Accordingly, the Norwegian meat industry might use CO as a component of a packaging gas in concentrations up to 0.5% until October 1, 2000. The high CO₂/low CO mixture is the only MAP which provides a shelf life sufficient for displaying and selling fresh retail meat products in all parts of Norway. The Norwegian meat industry started to use the high CO₂/low CO mixture in packaging of fresh retail meat products in the mid-eighties. The market share of retail meat packaged in the high CO₂/low CO mixture in Norway is currently estimated at 50 - 60% (ground beef as high as 85%). The Norwegian food control authority has not registered outbreaks or a higher frequency of sporadic cases of food borne diseases linked to such products (The Scientific Committee, under The Norwegian Food Control Authority, 19.4.99).

Support from the European meat industry:

The meat industry in Sweden has also discovered the benefits and advantages of the high CO₂/low CO mixture in packaging of fresh meat. Both Swedish Meats (which is the organisation of the Swedish meat cooperative) and the Swedish Meat Trade Association (which is the organisation of the private meat industry in Sweden) support the Norwegian meat industry's

application to the EU Commission (letters enclosed). Also, the Danish Pig Producers and Slaughterhouses, the Spanish Meat Industry and the Finnish Meat Research Institute support the application (letters enclosed).

- **benefit for the consumer:**

The high CO₂/low CO mixture is the only MAP which provides a shelf life sufficient for displaying and selling fresh retail meat products in a large geographical area like Norway.

Food safety and traceability:

The high CO₂/low CO mixture enables centralised packaging operations with quality control with less risk for cross-contamination than in local butcher shops or by supermarket back-store operations. The Norwegian meat industry already produces pork products traceable in integrated systems back to the farm and beef products traceable to the individual animal.

The ability of *Y. enterocolitica* to multiply at low temperature is of considerable concern to food producers, particularly in countries like Australia, Canada, Denmark, Germany, New Zealand, Norway and Sweden where *Y. enterocolitica* has surpassed *Shigella* and now rivals *Salmonella* and *Campylobacter* as a cause of acute bacterial gastroenteritis (Nesbakken, 2000). The growth of *Y. enterocolitica* was totally inhibited in ground beef packed in the high CO₂/low CO mixture both at 4°C and 10°C while it grew fairly well both in the high O₂ mixture and in the chub packs.

At the abusive storage temperature of 10°C *E. coli* O157:H7 in the chub packs grew about as fast as the background flora. However, growth was nearly totally inhibited in the high CO₂/low CO mixture and in the high O₂ mixture (Nissen et al., 1999; Nissen et al., submitted).

Quality:

Centralised pre-packaging of retail meat in the meat industry is cost-effective compared to on-site packaging in food stores. Self-service food stores and supermarkets often require to be supplied with pre-packaged meat. The long shelf life of meat packaged in the high CO₂/low CO mixture provides a possibility of a wider selection of fresh meat on display in the stores. If the Norwegian meat industry loses the possibility to use this mixture, food stores in rural and remote areas will have to be supplied by frozen meat, which has a low acceptability of the consumer.

The high CO₂/low CO mixture provides extended freshness: fresh meat will last for many days (often more than a week) in the consumer's home refrigerator, and the consumer might shop fresh meat once a week, and fresh meat is available 24 hrs a day 7 days a week in hypermarkets, supermarkets, discount stores, service stores; and the consumer might get quality premium brand fresh meat in her/his local discount store. The consumer will find a wider variety of fresh meat products than otherwise possible.

The consumers seem to prefer fresh meat products packaged in the high CO₂/low CO mixture or the high O₂ mixture compared to other packaging methods (Solheim 1996).

Leakages in packages containing high CO₂/low CO mixture might be detected by the consumer. The discoloration might be an indicator of leakages for ground beef packed in the high CO₂/low CO mixture (Sørheim, 1996).

Prices:

Industrialised handling with centralised packaging in MAP means lower consumer prices. Waste due to "sell by date" in the distribution chain in Norway (high CO₂/low CO mixture) is less than 1%, as compared to 2 - 3% in countries using the high O₂ mixture, according to interviews with operators/supermarket chains in UK, The Netherlands and Spain (Dag Hallan, Norwegian Meat Cooperative, personal communication).

II.6. Exposure

Carbon monoxide (CO) is a colourless, odourless and tasteless gas. It is produced by incomplete combustion of carbon-containing organic material. The production of CO from natural processes is quite significant. Nevertheless, CO from antropogenic activities is far more important concerning human health, since this formation takes place in heavily polluted areas.

Natural background levels of CO are 0.01 - 0.9 mg/m³ (0.01 - 0.8 ppm). In urban areas, 8-h mean concentrations of CO are generally < 20 mg/m³, but levels exceeding 60 mg/m³ have been reported (WHO, 1979). Among tobacco smokers, CO from smoking is by far the dominating source of CO exposure (WHO, 1987).

According to WHO experts (WHO, 1979; WHO, 1987), the only way of exposure which is of relevance to human health, is via inhalation of CO gas. Upon absorption from the lungs into the blood, CO forms a strong coordination bond with the iron atom in haemoglobin forming carboxyhaemoglobin (HbCO). The affinity of haemoglobin for CO is roughly 240 times that of its affinity for oxygen. CO is absorbed through the lungs and the concentration of HbCO in the blood will depend on several factors, mainly the concentration of CO in inhalation air, the exposure time and the level of activity of the individual (pulmonary ventilation).

The Norwegian meat industry is using a gas mixture of 60 - 70% CO₂, 30 - 40% N₂ and 0.3 - 0.4% CO for the packaging of fresh retail meat of beef, pork and lamb. According to Watts et al. (1978) beef which is exposed to an atmosphere containing 1% CO for 3 days result in about 30% saturation of the meat myoglobin. When the meat was cooked (hotplate maintained at 195°C for up to 8 minutes), only 0.1 mg CO remained in the meat per kg resulting in a loss of CO about 85%.

Data are very scarce concerning comparison of CO exposure from air and consumption of CO-treated meat. According to Sørheim et al. (1997a) consumption of 250 g CO-treated meat (after cooking) yield a theoretical intake of maximum 0.025 mg CO, compared with inhalation of 15 mg CO per hour at the acceptance level suggested by Norwegian authorities (giving 1.5% HbCO, including endogenous formation). Even though the estimates are crude, the calculations show without doubt that intake of CO from meat consumption is negligible. Furthermore, absorption of CO from the gastrointestinal tract will be very much lower (if it happens at all), compared with absorption via the lungs.

II.7. Reaction and fate in food

The main function of low levels of CO in MAs is to give a stable, cherry red colour of the meat through strong binding of CO to myoglobin and formation of carboxymyoglobin (El-Badawi, 1964). Although a substantial increase in the shelf life of meat can be obtained by using various MAs, it is often limited by discolouration due to oxidation of myoglobin to metmyoglobin. This discolouration can be prevented by including a small fraction of CO in the gas mixture. Carboxymyoglobin is more resistant to oxidation than oxymyoglobin, due to the stronger binding of CO to the iron-porphyrin site on the myoglobin molecule (Wolfe, 1980). CO in concentrations of 1 - 5% had the ability to increase metmyoglobin reduction, even in the presence of air (Lanier et al., 1978).

PART III. TOXICOLOGICAL DATA

III. 1 - 4.

Health effects of carbon monoxide has been evaluated by two WHO expert groups (WHO, 1979; WHO, 1987). The health effects are associated with the degree of HbCO formation. According to the aforementioned expert groups, the most sensitive individuals should be protected from CO exposures leading to a HbCO level exceeding 2.5%. In healthy adults, no adverse health effects are described at concentrations resulting in HbCO levels < 5%.

A small amount of CO is formed naturally in the human body, from breakdown of haemoproteins. This production leads to a HbCO concentration of about 0.5%. The average HbCO concentration in non-smokers is 1.2 - 1.5%, while the level in smokers usually is 3 - 4%.

The WHO experts (WHO, 1987) recommended a maximum HbCO level of 2.5 - 3% in order to protect the population at large, included sensitive individuals. In order to obtain this, they recommended maximum levels of CO in ambient air which will meet this requirement for different times of exposure:

- Maximum levels of 100 mg/m³ for < 15 min
- Average levels < 60 mg/m³ for 30 min
- Average levels < 30 mg/m³ for 1 hour
- Average levels < 10 mg/m³ for 8 hours

The European Union has not evaluated CO for use as a packaging gas for meat. However, in 1990 (European Commission, 1991), several other gases (carbon dioxide, oxygen, nitrogen, nitrous oxide, hydrogen and argon) were evaluated by the Scientific Committee for Food (SCF) for use as packaging gases and propellants. In this case it was considered unnecessary to adopt ADIs because of general knowledge of their safety in use, and the estimated insignificant intakes compared with exposure from other sources. Furthermore, in 1996, the SCF reviewed the safety of modified and controlled atmosphere packaging again, and placed particular emphasis on the importance of HACCP for the avoidance of microbiological risk in this context (European Commission, 1996). The SCF concluded that it does not see specific hazards for human health by

the use of controlled or MAs, but that a prerequisite is that the principles of HACCP are observed. A similar approach should also be feasible concerning CO used at very low concentrations in mixture with CO₂ and N₂.

Accordingly, the issue which should be solved concerning health effects of CO used in gas packaging, is the question of preventing the consumers from exposure to meat of unacceptable microbiological quality. Thus, two studies on shelf life, off-odour and colour (Sørheim et al., 1999) and pathogens (Nissen et al., 1999; Nissen et al., submitted) follow as enclosures. Summaries of the two studies are also given in "Part II.5. Justification for the additive - investigations on the efficacy of the substance for the intended effect at the level proposed".

III.5. Review of results and conclusions

As can be seen from the foregoing, exposure to CO via consumption of meat products treated with a MA containing < 0.5% CO represent a negligible source of CO, and will probably not contribute to any increase in the carboxyhaemoglobin level. From a toxicological point of view, packaging gas with < 0.5% CO presents no threat to human health (Sørheim et al. 1997a). This is in accordance with an assessment performed by members of The Scientific Committee for Food, under The Norwegian Food Control Authority (30.11.98).

PART IV. SUMMARY DOCUMENT

Gas mixtures with low concentrations of CO and high concentrations of CO₂ provide a combination of a long microbiological shelf life and a stable bright red colour of meat. Meat packaged in a MA with high O₂ can achieve an initial bright red colour, but the microbiological shelf life and the colour stability are considerably lower than those of the CO mixture. Using CO eliminates the need for having O₂ as a component of the MA. Other MAs and packaging methods, like high CO₂ with O₂ absorbers, chub packs and vacuum packs may give a microbiological shelf life similar to that of the high CO₂/low CO mixture, but with a less acceptable colour or appearance of the meat. Thus, there appears at present to be no fully satisfactory alternatives to the CO mixture used in packaging of retail-ready red meats in Norway (Sørheim et al., 1999).

In an investigation, growth of *E. coli* O157:H7 at 10°C in ground beef was nearly totally inhibited in the high CO₂/low CO mixture. The prolonged shelf life at 4°C did not increase growth of *L. monocytogenes* in ground beef stored in the high CO₂/low CO mixture. The growth of *Y. enterocolitica* was totally inhibited in ground beef packed in the high CO₂/low CO mixture both at 4°C and 10°C while it grew fairly well both in the high O₂ mixture and in the chub packs. However, the observed growth of strains of *Salmonella* both in the high CO₂/low CO mixture and in chub packs at the abuse temperature of 10°C does emphasise the importance of temperature control during storage (Nissen et al., 1999; Nissen et al., submitted).

From a toxicological point of view, packaging gas with < 0.5% CO presents no threat to human health (Sørheim et al. 1997a). The European Union has not evaluated CO for use as a packaging gas for meat. However, in 1990 (European Commission, 1991), several other gases (carbon dioxide, oxygen, nitrogen, nitrous oxide, hydrogen and argon) were evaluated by the Scientific

Committee for Food (SCF) for use as packaging gases and propellants. In this case it was considered unnecessary to adopt ADIs because of general knowledge of their safety in use, and the estimated insignificant intakes compared with exposure from other sources. A similar approach should also be feasible concerning CO used at very low concentrations in mixture with CO₂ and N₂.

The Norwegian meat industry started to use the high CO₂/low CO mixture in packaging of fresh retail meat products in the mid-eighties. The market share of retail meat packaged in the high CO₂/low CO mixture in Norway is currently estimated at 50 - 60% (ground beef as high as 85%). The Norwegian food control authority has not registered outbreaks or a higher frequency of sporadic cases of food borne diseases linked to such products (The Scientific Committee, under The Norwegian Food Control Authority, 19.4.99).

Conclusions:

Gas mixtures with low concentrations of CO, up to 0.5%, and high levels of CO₂, approximately 70%, have many advantages regarding shelf life, inhibition of pathogenic bacteria like *E. coli* O157 and *Y. enterocolitica*, colour stability, labour safety and costs. CO used as described in these concentrations, does not present any toxic threat to the consumer. Considering the benefits the Norwegian meat industry has experienced with the CO gas mixture over the past decade, this gas mixture should have a potential for a wider application in retail packaging of meat in the EU.

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1 PACKAGING OF GROUND BEEF IN AN ATMOSPHERE WITH HIGH
2 CARBON DIOXIDE AND LOW CARBON MONOXIDE RESTRAINS
3 GROWTH OF *YERSINIA ENTEROCOLITICA*, *LISTERIA*
4 *MONOCYTOGENES* AND *ESCHERICHIA COLI* O157:H7

5

6 H. Nissen^{*}, O. Alvseike^{1,2}, S. Bredholt, A. Holck and T. Nesbakken²

7

8 MATFORSK, Norwegian Food Research Institute, Oslovn. 1, N-1430 Ås, Norway

9

10 ¹The Norwegian School of Veterinary Science, P.O. Box 8146 Dep., 0033 Oslo,
11 Norway

12 ²Norwegian Meat Research Centre, P.O. Box 396, 0513 Oslo, Norway

13

14 Corresponding author and request for reprints: Hilde Nissen, MATFORSK,
15 Norwegian Food Research Institute, Oslovn. 1, N-1430 Ås, Norway

16 Tel: +47 64970100

17 Fax: +47 64970333

18 E-mail: hilde.nissen@matforsk.no

1 Abstract

2 Growth of the pathogens *Yersinia enterocolitica*, *Listeria monocytogenes*,
3 *Escherichia coli* O157:H7 and strains of *Salmonella* was compared in ground beef
4 packed in modified atmospheres of 60 % CO₂/ 40 % N₂ /0.4 % CO (high CO₂/ low
5 CO mixture), 70 % O₂/ 30 % CO₂ (high O₂ mixture) and in chub packs. The ground
6 beef was inoculated with rifampicin-resistant or nalidixic acid/streptomycin-resistant
7 strains (final concentration 10²-10³ bacteria/g) and stored at 4 and 10 °C for up to 14
8 days. At 4 °C the shelf life based on stable colour and reduced background flora was
9 prolonged for the high CO₂/ low CO mixture compared to the two other packaging
10 methods, but at 10 °C the shelf life was < 8 days for all the packaging methods.
11 Growth of *Y. enterocolitica* was nearly totally inhibited both at 4 and 10 °C in the high
12 CO₂/ low CO mixture, while the bacterial numbers in the samples packed in the high
13 O₂ mixture increased from about 5x10² bacteria/g at day 0 to about 10⁴ at day 5 at
14 4°C and to 10⁵ at 10°C. Growth in the chub packs was even higher. *Listeria*
15 *monocytogenes* showed very little growth at 4 °C in all treatments. At 10 °C there
16 was slow growth from about 5x10³ bacteria/g to about 10⁴ at day 5 in the high CO₂/
17 low CO mixture, while the numbers in the high O₂ mixture and the chub packs were
18 about 10 times higher. Growth of *E. coli* O157:H7 at 10 °C in the ground beef was
19 nearly totally inhibited in both the high CO₂/ low CO mixture and the high O₂ mixture.
20 Growth in the chub packs was higher, reaching 10⁵ bacteria/g on day 5. The
21 *Salmonella* strains (*S. Typhimurium*, *S. Dublin*, *S. Enteritidis* and *S. enterica*
22 61:k:1,5,(7)) in the ground meat stored at 10 °C for 5 and 7 days grew to a higher
23 number in the high CO₂/ low CO mixture than in the high O₂ mixture. This study
24 shows that the prolonged shelf life at 4 °C did not increase growth of *Y. enterocolitica*

1 and *L. monocytogenes* in ground beef stored in the high CO₂/ low CO mixture
2 mixture, but the observed growth of strains of salmonella at 10 °C in this mixture and
3 in chub packs does emphasise the importance of temperature control during storage.

4
5 **Keywords:**

6 Ground beef, modified atmosphere packaging, high CO₂, carbon
7 monoxide, *Yersinia enterocolitica*, *Listeria monocytogenes*, *Escherichia coli*
8 O157:H7.

9

1. Introduction

Ground beef for retail sale is most often ready-packed in modified atmospheres (MA) or in chub packs. MA-packed ground beef has a longer microbiological shelf life and also maintains an attractive red colour. For the past decade the Norwegian meat industry has been using a gas mixture of 60-70 % CO₂, 30-40 % N₂, 0.3-0.5 % CO. (The CO comes ready mixed in the N₂ from the supplier.) The reason for adding CO to the gas mixture is that it will produce a long-lasting cherry-red colour of the meat (Sørheim et al., 1999), but the low concentration of CO has little effect on the microflora of the meat (Clark et al., 1976; Gee and Brown, 1978; Luno et al., 1998). The use of CO at such low concentrations does not present any toxic threat to the consumers (Sørheim et al., 1997). The most commonly used gas mixture for retail-ready meat in other European countries is 70 % O₂/30 % CO₂ (Gill, 1996). The high oxygen concentration is needed to keep the red colour of the meat (Lambert et al., 1991). It is therefore only possible to obtain half the CO₂ concentration used in the high CO₂/ low CO mixture. The microbiological shelf life of the high O₂ mixture will be longer than in air, but less than in the high CO₂/ low CO gas mixture (Sørheim et al., 1999).

The inclusion of CO is controversial because the stable cherry-red colour can last beyond the microbiological shelf life of the meat and thus mask spoilage (Kropf, 1980). The extended shelf life obtained by MAP may under some conditions imply increased risk of growth of pathogens (Silliker and Wolfe, 1980; Hintlian and Hotchkiss, 1986; Farber, 1991; Lamberts et al., 1991). This issue has also been discussed by the European Commission (1997).

However, even if meat packed in high CO₂/ low CO mixture acquires a stable colour, the shelf life based on odour is significantly longer in the high CO₂/ low CO

1 mixture only at 4 °C (Sørheim et al., 1999). At this temperature *Yersinia enterocolitica*
2 and *Listeria monocytogenes* are considered to be the most serious pathogens in
3 meat. At abuse temperatures (>8 °C) *Escherichia coli* O157:H7 and *Salmonella* spp.
4 also may grow and increase the health risk to the consumers. In the present study
5 we wanted to compare growth of these pathogens in ground beef packed in a
6 commercial Norwegian 60 % CO₂/40 % N₂/0.4 % CO (high CO₂/low CO mixture) with
7 growth in a high O₂ (70 % O₂/30 % CO₂) gas mixture and in ground beef in chub
8 packs during storage at 4 and 10 °C in order to evaluate the microbiological safety of
9 the product.

10

11 2. Materials and methods

12 2.1. Preparation and packaging of the ground beef

13 The beef carcasses were de-boned, and trimmings with 14 % fat were ground
14 through a 4 mm plate. The batch of ground beef was divided into 500 g portions
15 which were packaged in 0.4 % CO/ 60 % CO₂/ 40 % N₂ (high CO₂/ low CO mixture),
16 70 % O₂/ 30 % CO₂ (high O₂) or packed in clipped chub packs. The beef was packed
17 at a commercial meat plant within 1 hour of grinding as described by Sørheim et al.
18 (1999).

19

20 2.2. Bacterial cultures and growth conditions

21 Strains of the following pathogens were inoculated in the ground beef: *Yersinia*
22 *enterocolitica* (mixture of 3 strains), *Listeria monocytogenes* (mixture of 3 strains
23 isolated from cooked sausage, Blom et al., 1997, Nissen and Holck, 1999),
24 *Escherichia coli* O157:H7, NCTC 1200 (National Collection of Type Cultures,

1 Colindale, London), non-toxic strain (resistant to 100 µg/ml nalidixic acid and 1000
2 µg/ml streptomycin) and *Salmonella enterica* subspecies *diarizonae* serovar
3 61:k:1,5,(7) (*S. enterica* 61:k:1,5,(7)), mixture of 3 strains (National Institute of Public
4 Health, Oslo). The listeria and yersinia strains were made resistant to rifampicin by
5 spreading 0.1 ml of overnight cultures onto agar plates of TSB medium (Oxoid, CM
6 129) containing 50 µg/ml rifampicin (Sigma, St.Louis, MO, USA). The growth rates of
7 the resistant strains were practically equal to those of the parent strains when tested
8 in TSB medium in a Bioscreen Instrument (Labsystem Co., Helsinki, Finland) at the
9 same temperature, pH and a_w (NaCl) concentrations.

10 In a second experiment four rifampicin-resistant salmonella strains, *S.*
11 *Typhimurium*, *S. Dublin*, *S. Enteritidis* and *S. enterica* 61:k:1,5,(7) were used to
12 inoculate the MAP- packed ground beef. The growth rates (measured as above) of
13 the resistant strains of *S. Enteritidis* and *S. enterica* 61:k:1,5,(7) were essentially the
14 same as the parent strains while the growth rates of *S. Dublin* and *S. Typhimurium*
15 were slightly lower.

16 17 2.3. Inoculation and storage

18 After packaging the ground beef was inoculated with stationary cultures (the
19 bacteria were cultivated overnight at 30°C and kept in the refrigerator for 1 day
20 before use) of the different pathogenic bacteria. The stock cultures were diluted in
21 peptone water (PW) (Bacto peptone, Difco, 1g/l; NaCl, Merck, 8.5 g/l) and the strains
22 belonging to the same species or serovars were mixed. Fifty µl of each pathogen
23 were inoculated with a syringe through a gas probe self-sealing tape (Toray
24 Engineering Co. Ltd, England) into one of the corners of the MA packages. The
25 packages thus had one pathogen inoculated in each corner. In the chub packs the

1 pathogens were inoculated at least 3 cm apart. Packages inoculated only with *Y.*
2 *enterocolitica* and *L. monocytogenes* only were stored at 4°C and analysed after 0, 2,
3 5, 8 and 14 days while packages inoculated with all 4 pathogens were stored at
4 10 °C and analysed after 0, 2, 5 and 8 days.

5 In the second experiment four serovars of «*Salmonella*» were inoculated in one
6 corner each of the package of ground beef and which was stored at 10 °C and
7 analysed after 0, 2, 5 and 7 days. Non-inoculated packages used as controls were
8 also stored at 10 °C.

9 10 2.4. Microbial analyses

11 Samples of 25 g ground beef containing the inoculated pathogens were
12 transferred to a stomacher bag and mixed with 150 ml peptone water (8.5 g NaCl,
13 1.0 g peptone/1000 ml water). One hundred µl of a ten-fold dilution series were
14 plated on blood agar containing 50 µg/ml rifampicin for *L. monocytogenes* and *Y.*
15 *enterocolitica* or 100 µg/ml nalidixic acid and 1000 µg/ml streptomycin sulphate for *E.*
16 *coli* O157:H7. From the undiluted mixture an aliquot of 1 ml was also plated out. For
17 enumeration of *Salmonella* spp. the selective medium Brilliant Green Agar (modified)
18 (BGA; Oxoid, Basingstoke, Hampshire, England) was used. The colonies were
19 confirmed on Triple Sugar Iron Agar (TSI; Difco, Detroit, MI,) and Urea agar (Urea
20 Agar Base, Oxoid CM53 and Urea Solution, Oxoid SR20) followed by agglutination
21 by monovalent antisera (provided by the National Institute of Public Health). In the
22 second experiment, samples for detection of the four salmonella strains were plated
23 on blood agar containing 50 µg/ml rifampicin samples from non-inoculated packages
24 were treated the same way and plated on MRS plates (CM359, Oxoid), pH 5.7, for
25 determination of lactic acid bacteria and PCA (Difco, Detroit, MI, USA) plates for total

1 counts of bacteria. The plates were incubated at 30°C for up to 2 days, all
2 aerobically. On each sampling date the packs with MA were analysed for O₂ and CO₂,
3 and the pH for all samples was measured in the stomacher solution. Samples from
4 two replicate packages were used for all analyses, except after 7 days storage in
5 experiment 2 where three replicate packages were analysed.

7 2.5. Statistical analyses

8 Microbial data were subjected to analysis of variance (ANOVA) and Tukey's
9 pairwise comparisons. It was deemed appropriate to perform ANOVA on these data
10 after a log₁₀ transformation, thereby obtaining a distribution more akin to the normal
11 distribution on which ANOVA is based.

13 3. Results

14 As expected the shelf life of the ground beef stored at 4 °C was prolonged in the
15 high CO₂/ low CO mixture compared with the other packaging methods. This was due
16 to the stable colour and reduced background flora resulting in little off-odour.

17 Thus the ground beef packed in the high CO₂/ low CO mixture still had an acceptable
18 smell after 14 days of storage at 4 °C, while the beef packed in high O₂ mixture and
19 in the chub packs had some off-odours. The difference in shelf life was less at 10 °C.

20 After 5 days storage the ground beef packed in the high CO₂/ low CO mixture had an
21 acceptable smell (except the packages inoculated with salmonella, while beef packed
22 in the high O₂ mixture and the chub packs had a slight off-odour.

23 After 8 days storage there was a strong off-odour for all treatments, but the ground
24 beef in the high CO₂/ low CO mixture still looked bright red, in accordance with

25 Sørheim et al. (1999). The O₂ content in the high CO₂/ low CO mixture was virtually

1 zero throughout storage at both temperatures. At 10 °C the O₂ content in the high O₂
2 gas mixture decreased from 70 to about 35 % after 8 days storage, probably due to
3 aerobic bacterial metabolism. The chub packs had an O₂-permeable casing which
4 probably was the cause of the high bacterial growth in these packs at both
5 temperatures.

6 Growth of *Y. enterocolitica* was totally inhibited both at 4 and 10 °C in the high
7 CO₂/ low CO mixture (Fig. 1a and b), while the number in the samples packed in the
8 high O₂ mixture increased from about 5x10² cfu/g at day 0 to about 10⁴ cfu/g at day 5
9 at 4 °C and to 10⁵ cfu/g at 10°C. Growth in the chub packs at 4 °C was even higher
10 than in the other treatments. Growth in chub packs was also higher than in high O₂ at
11 10 °C (p=0.007). *L. monocytogens* (Fig. 2a) showed very little growth at 4 °C in all
12 treatments. At 10 °C (Fig. 2b) there was slow growth (from about 5x10³ bacteria/g to
13 about 10⁴ at day 5) in the high CO₂/ low CO mixture. This was more than 10-fold
14 higher cfu/g at day 5 than in the high O₂ mixture (p= 0.040) and the chub packs
15 (p=0.035). Ground beef inoculated with *E. coli* O157:H7 and strains of salmonella
16 was stored at 10°C. Growth of *E. coli* O157:H7 was slow both in the high CO₂/ low
17 CO mixture and the high O₂ mixture (Fig. 3) and the numbers were less than 10⁴
18 cfu/g at day 5. Growth in the chub packs was greater than in the high CO₂/ low CO-
19 mixture (p=0.011) and in the high O₂ mixture (p=0.019), reaching 10⁵ cfu/g. Growth of
20 lactic acid bacteria in the non-inoculated packages was somewhat inhibited in the
21 high CO₂/ low CO mixture, especially at 4 °C (Fig. 4). At start of the experiment the
22 pH in the ground beef was about 5.8 in all packages. After 5 days storage the pH
23 was about 5.7 in the high CO₂/ low CO mixture, 5.5 in the high O₂ mixture and 5.3 in
24 the chub packs.

1 Due to growth of other bacteria on the selective plates, only approximate numbers
2 of *S. enterica* 61:k:1,5,(7) were obtained, but growth of about 1.5 log units was
3 observed both in the CO mixture and the chub packs (results not shown). This
4 increase was not seen in the high O₂ mixture. To verify these results and check
5 whether they were valid for other serovars more virulent to humans, such as *S.*
6 *Typhimurium*, *S. Dublin* and *S. Enteritidis*, a second experiment was performed. The
7 results (Fig. 5 a, b, c and d) show that after 2 days of storage at 10 °C there was
8 essentially no growth of the salmonella strains in ground beef packed in the high
9 CO₂/ low CO mixture and in the high O₂ mixture, while the numbers of salmonella in
10 the chub packs were about 10 fold higher. After 5 days there was a slight off-odour in
11 all the packages except for one package with high CO₂/ low CO mixture which
12 smelled strongly of H₂S. In this package the numbers of all the salmonella strains
13 were higher than in the replicate package and were of the same magnitude as the
14 numbers in the chub packs. In the O₂ mixture there was no growth of *S. Dublin* and
15 *S. Enteritidis* and only a low growth of *S. enterica* 61:k:1,5,(7) and *S. Typhimurium*.
16 The growth of the salmonella strains was still greatly inhibited in the high O₂ mixture,
17 while growth in the high CO₂/ low CO mixture was just as high or even higher than in
18 the chub packs.

19 In the non-inoculated packages the lactic acid bacteria rapidly constituted most of
20 the background flora (not shown). After 5 days storage the numbers were higher in
21 the chub-packed samples, but after 8 days there were no obvious differences
22 (Fig. 6). The pH in the non-inoculated ground beef followed the same pattern as in
23 experiment 1.

1 4. Discussion and Conclusions

2 Ground beef is a high-risk product because pathogens may be mixed into the
3 ground product which may not be sufficiently heated before consumption. To inhibit
4 growth of spoilage bacteria and increase shelf life, MAP is often used by retailers.
5 The question «Do modified atmospheres enhance risk to the consumers health, but
6 delay signs of spoilage» raised by Hintlian and Hotchkiss (1986) is therefore relevant.
7 When evaluating the safety of ground beef in the high CO₂/ low CO mixture
8 compared to other commercially available packaging methods, we have focused on
9 bacteria that show good growth below 10 °C and are most relevant for meat
10 products.

11 The ability of *Y. enterocolitica* to multiply at low temperature is of considerable
12 concern to food producers, particularly in countries like Australia, Canada, Denmark,
13 Germany, New Zealand, Norway and Sweden where *Y. enterocolitica* has surpassed
14 *Shigella* and now rivals *Salmonella* and *Campylobacter* as a cause of acute bacterial
15 gastroenteritis (Nesbakken, 1999). In our study, growth of *Yersinia enterocolitica* was
16 totally inhibited in ground beef packed in the high CO₂/ low CO mixture even at 10 °C
17 while it grew fairly well both in the high O₂ mixture and in the chub packs. Manui-
18 Tawiah et al. (1993) found that pork shops packed in different MA with 20 or 40 %
19 CO₂ with or without O₂ allowed growth of *Yersinia enterocolitica*, but here the CO₂
20 concentration was lower than in the high CO₂/ low CO mixture (60 %) used in our
21 study.

22 *Listeria monocytogenes* is also a pathogen that grows well at low temperatures,
23 but in our study there was no growth of this bacterium in the ground beef in any of
24 the packages at 4 °C, and only slow growth at 10 °C. This agrees with results of

1 Farber and Daley (1994) who found no growth of *L. monocytogenes* in different meat
2 products when stored at 4 °C.

3 At the abusive storage temperature of 10 °C, *E. coli* O157:H7 in the chub packs
4 grew about as fast as the background flora. However, growth was nearly totally
5 inhibited in the high CO₂/ low CO mixture and in the high O₂ mixture. This is in
6 accordance with the predictive model of Sutherland et al. (1997). Their study showed
7 that *E. coli* O157:H7 is relatively tolerant of CO₂, but growth could be inhibited at
8 10 °C at high CO₂ concentrations and pH < 6.0.

9 In our study, growth of *Salmonella* spp. was not inhibited in ground beef packed in
10 high CO₂/ low CO mixture and stored at 10 °C, contrary to what is found in many
11 other studies (e.g. D'Aoust, 1991). Although salmonella may grow well and out-
12 compete the background flora on fresh meat stored at 10 °C (Alford and Palumbo,
13 1969; Mackey and Kerridge, 1988), most reports claim that growth will be inhibited in
14 MAP at this temperature (Siliker and Wolfe, 1980; D'Aoust, 1991; Gill and DeLacy,
15 1991). Nychas and Tasson (1996) found that high CO₂ atmospheres were more
16 inhibitory for growth of *S. Enteritidis* on fresh poultry at 10 °C than were high O₂
17 atmospheres, the opposite of what we found for ground beef. Inhibition of bacterial
18 growth may, however, be influenced by pH, texture and the composition of the
19 product, and Gill and DeLacy (1991) did find growth of *S. Typhimurium* in high-pH
20 beef packed in CO₂ and stored at 10 °C. Oxidative stress reactions in salmonella
21 have recently been reported (Stephen et al., 1999). This may explain the inhibition of
22 growth (longer lag phase) in the high O₂ mixture in our study.

23 The present study shows that the prolonged shelf life (due to stable colour and
24 reduced background flora) at 4 °C did not increase the risk of growth of *Y.*
25 *enterocolitica* and *L. monocytogenes* in ground beef stored in the high CO₂/ low CO

1 gas mixture. This is probably due to the high CO₂ concentration that is inhibitory to
2 most microorganisms (Dixon and Kell, 1989). Even at the abusive temperature of
3 10 °C, the numbers of pathogens at the end of the shelf life (5 days) were less or the
4 same as were found in the chub packs. The observed growth of salmonella in the CO
5 mixture and chub packs does however emphasise the importance of temperature
6 control during storage. There is a wide range of temperature criteria for chilled foods
7 at retail in European countries. The values range from -1 °C to 10 °C, with most
8 temperatures being between 4 and 8 °C (European Commission, 1996). These
9 aspects should also be considered together with the conclusions of the EU report
10 (European Commission, 1997) which state that MAP has proven to enhance the
11 product quality by inhibiting the spoilage bacteria. MAP may also constitute a hurdle
12 to the growth of some pathogens, and the safety of MAP products are mostly
13 threatened by temperature abuse.

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1 Fig. 1. Growth of *Yersinia enterocolitica* inoculated in ground beef packed in high
2 CO₂/ low CO mixture (0.4 % CO/ 60 % CO₂/ 40 % N₂), high O₂ (70 % O₂/ 30 % CO₂)
3 or in chub packs. The ground beef was stored at a, 4 °C or b, 10 °C.

4

5 Fig. 2. Growth of *Listeria monocytogenes* inoculated in ground beef packed in high
6 CO₂/ low CO mixture (0.4 % CO/ 60 % CO₂/ 40 % N₂), high O₂ (70 % O₂/ 30 % CO₂)
7 or in chub packs. The ground beef was stored at a, 4 °C or b, 10 °C.

8

9 Fig. 3. Growth of *Escherichia coli* O157: H7 inoculated in ground beef packed in high
10 CO₂/ low CO mixture (0.4 % CO/ 60 % CO₂/ 40 % N₂), high O₂ (70 % O₂/ 30 % CO₂)
11 or in chub packs, stored at 10 °C.

12

13 Fig. 4. Growth of lactic acid bacteria (cfu/g on MRS, pH 5.7) in non-inoculated ground
14 beef packed in high CO₂/ low CO mixture (0.4 % CO/ 60 % CO₂/ 40 % N₂), high O₂
15 (70 % O₂/ 30 % CO₂) or in chub packs. The ground beef was stored at a, 4°C or b, 10
16 °C.

17

18 Fig. 5. Growth of strains of *Salmonellae* inoculated in ground beef packed in high
19 CO₂/ low CO mixture (0.4 % CO/ 60 % CO₂/ 40 % N₂), high O₂ (70 % O₂/ 30 % CO₂)
20 or in chub packs, stored 10 °C. a. *S. Typhimurium* b. *S. Dublin* c. *S. Enteritidis* d. *S.*
21 *enterica* 61:k:1,5,(7).

22

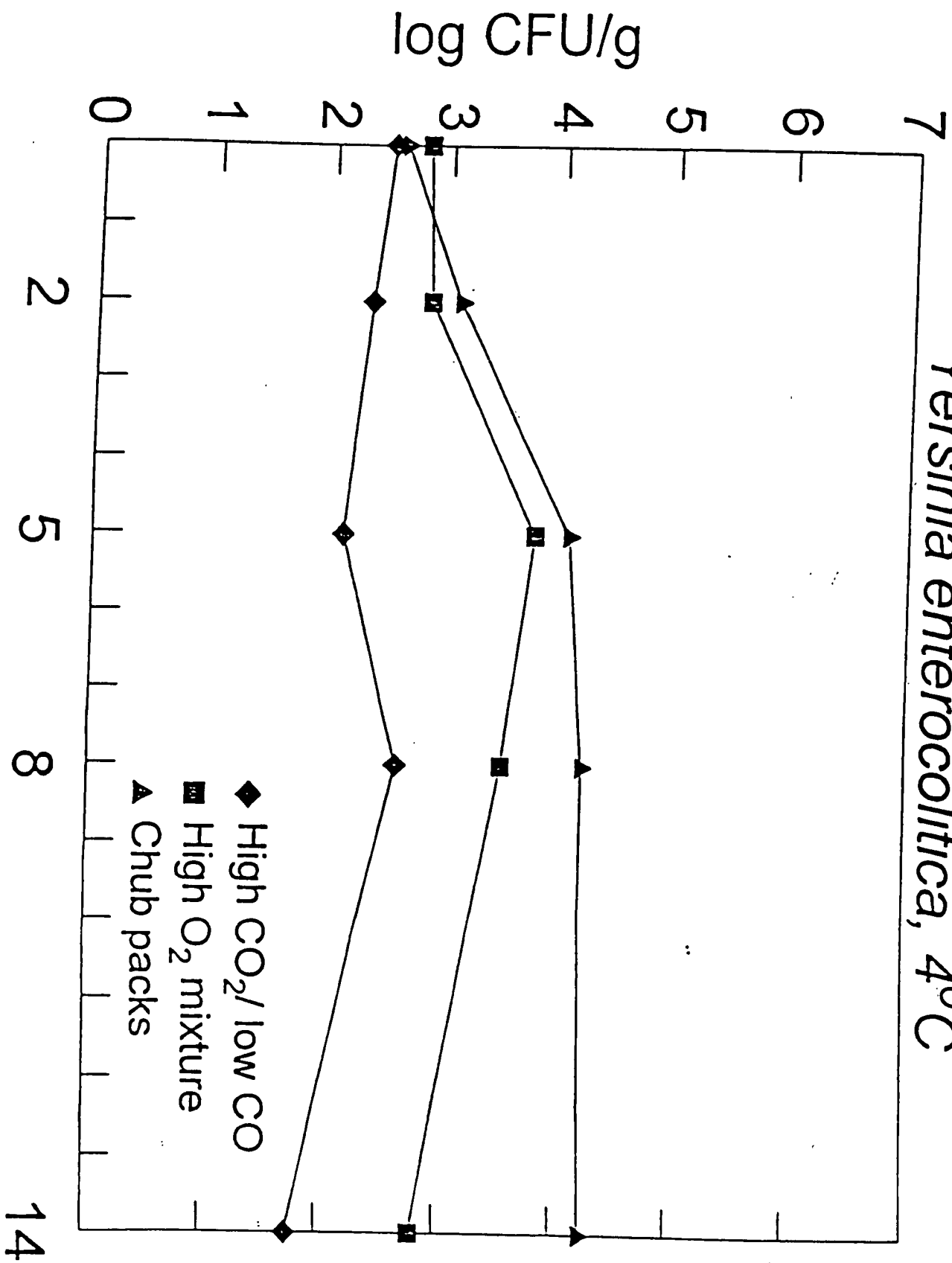
23 Fig. 6. Growth of lactic acid bacteria (cfu/g on MRS, pH 5.7) in non-inoculated ground
24 beef packed in high CO₂/ low CO mixture (0.4 % CO/ 60 % CO₂/ 40 % N₂), high O₂

1 (70 % O₂/ 30 % CO₂) or in chub packs. The ground beef was stored at a, 4 °C or b,
2 10 °C.

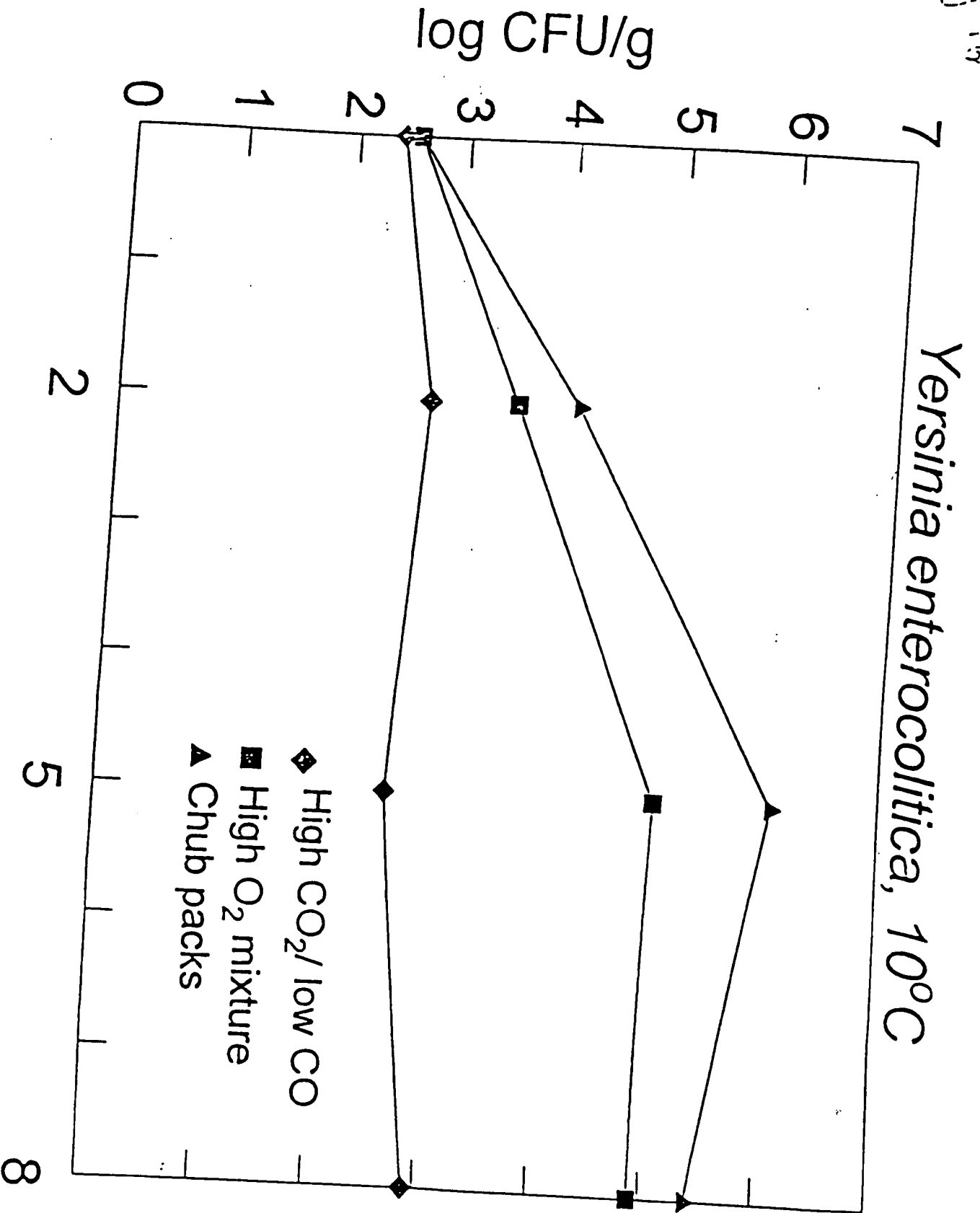
3

log. ha

Yersinia enterocolitica, 4°C



log



Listeria monocytogenes, 4°C

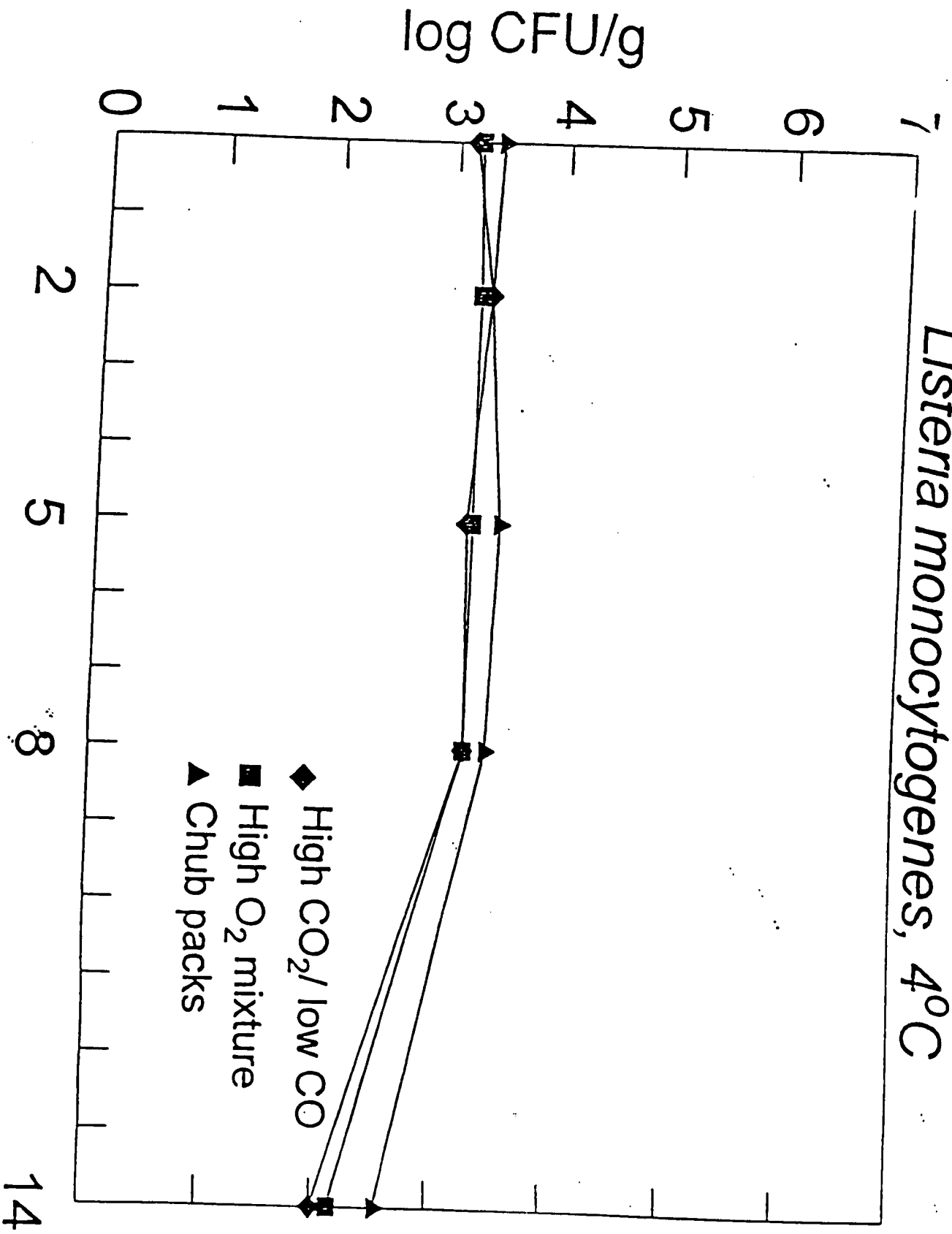


Fig. 2b

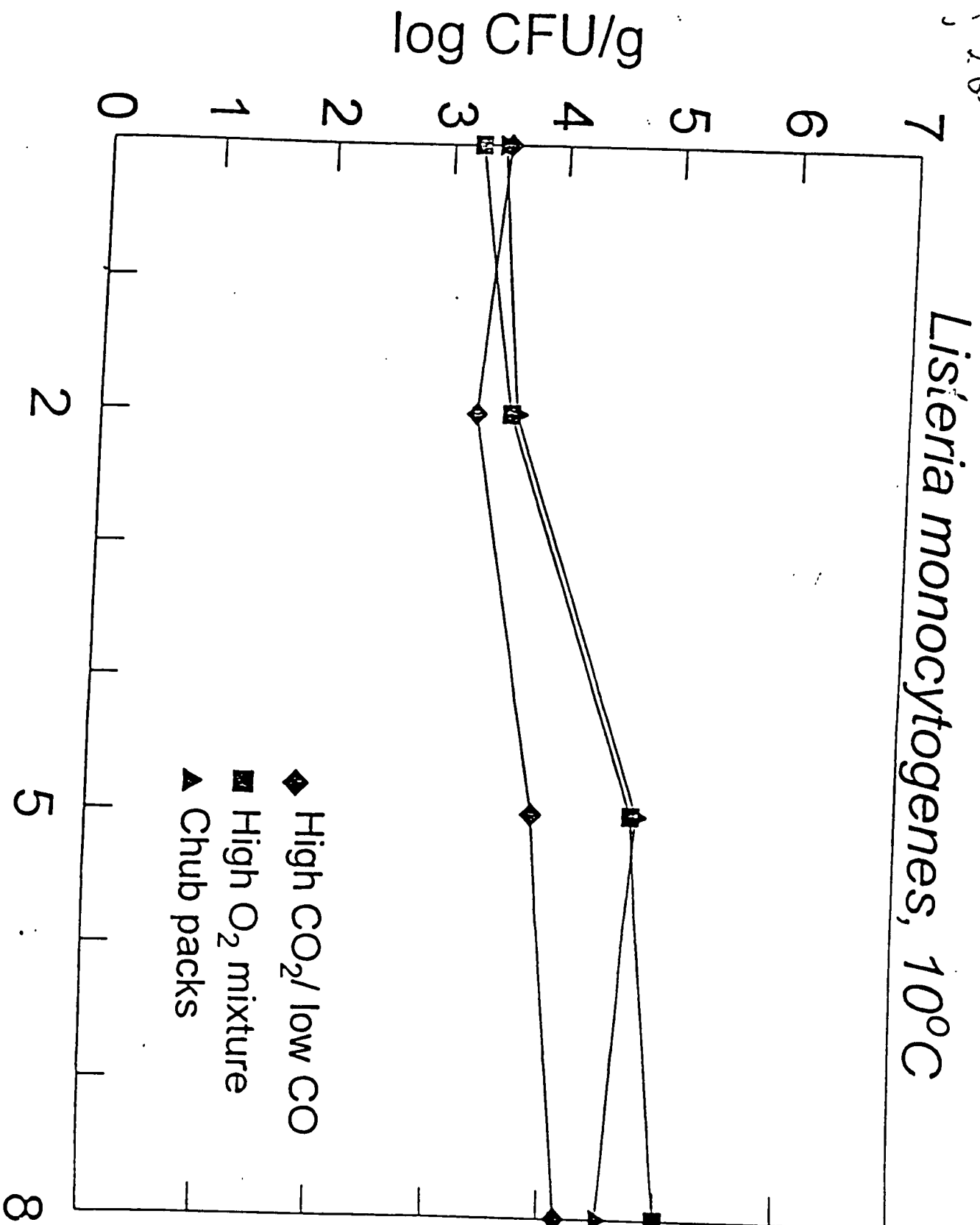
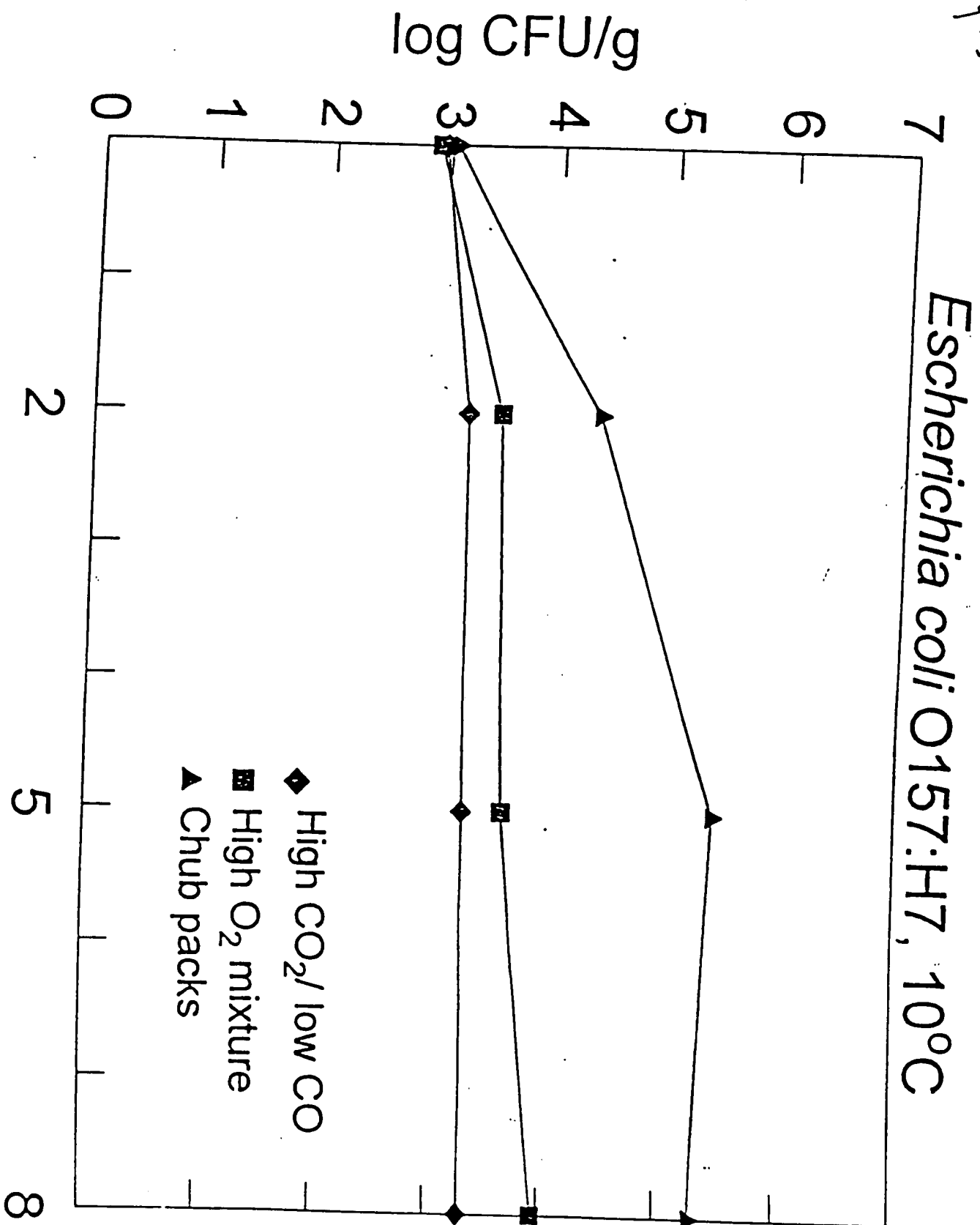


Fig. 3



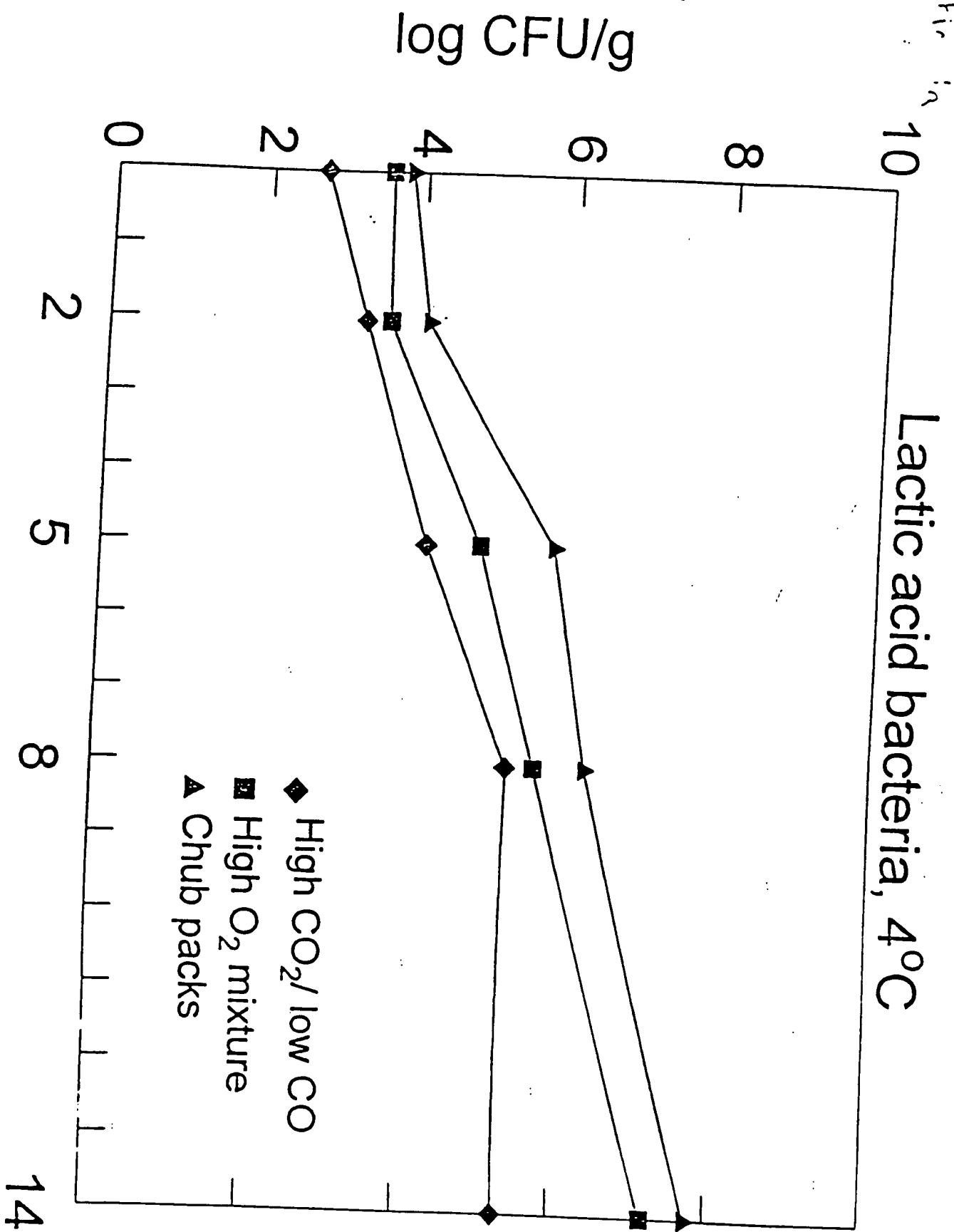


Fig. 16

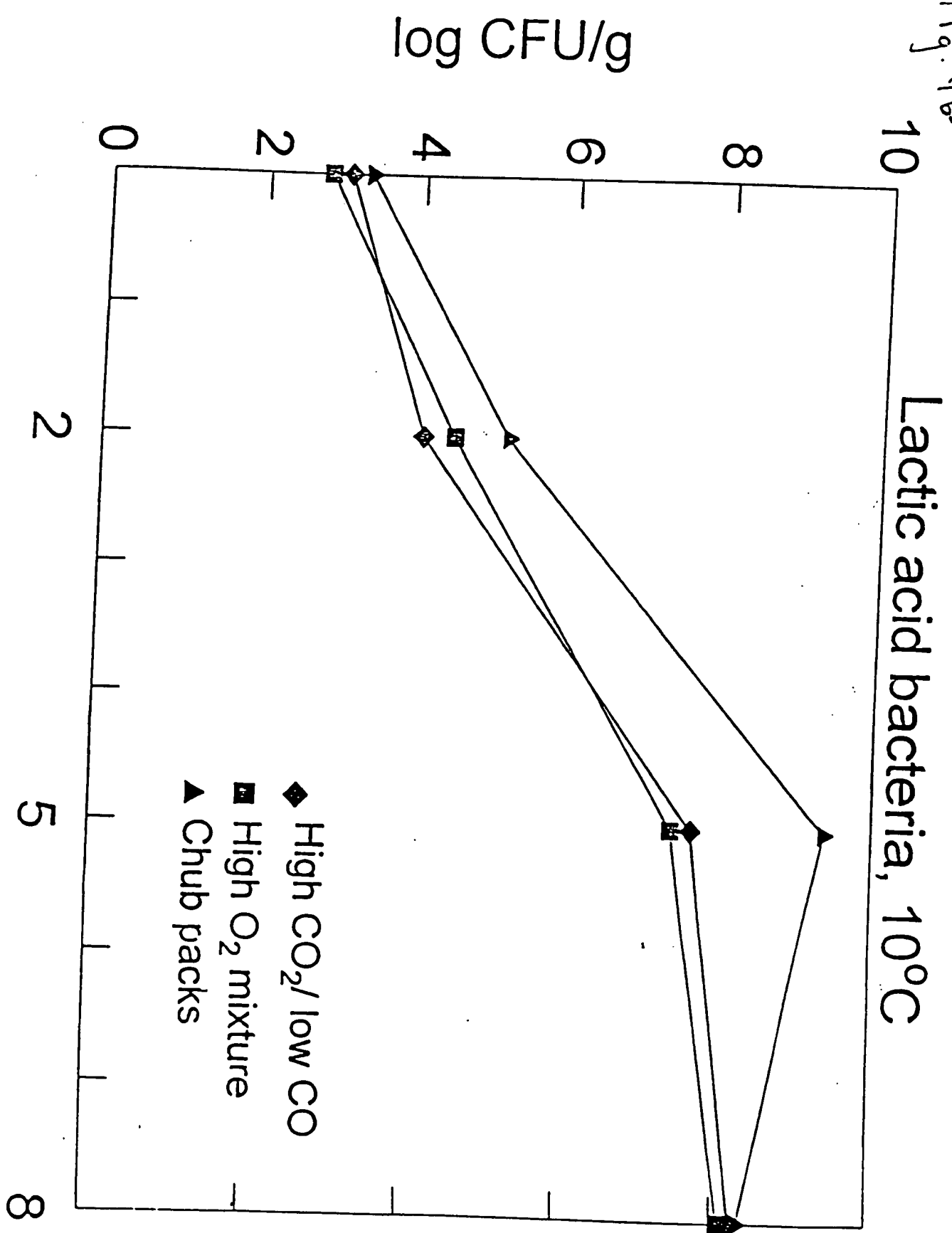
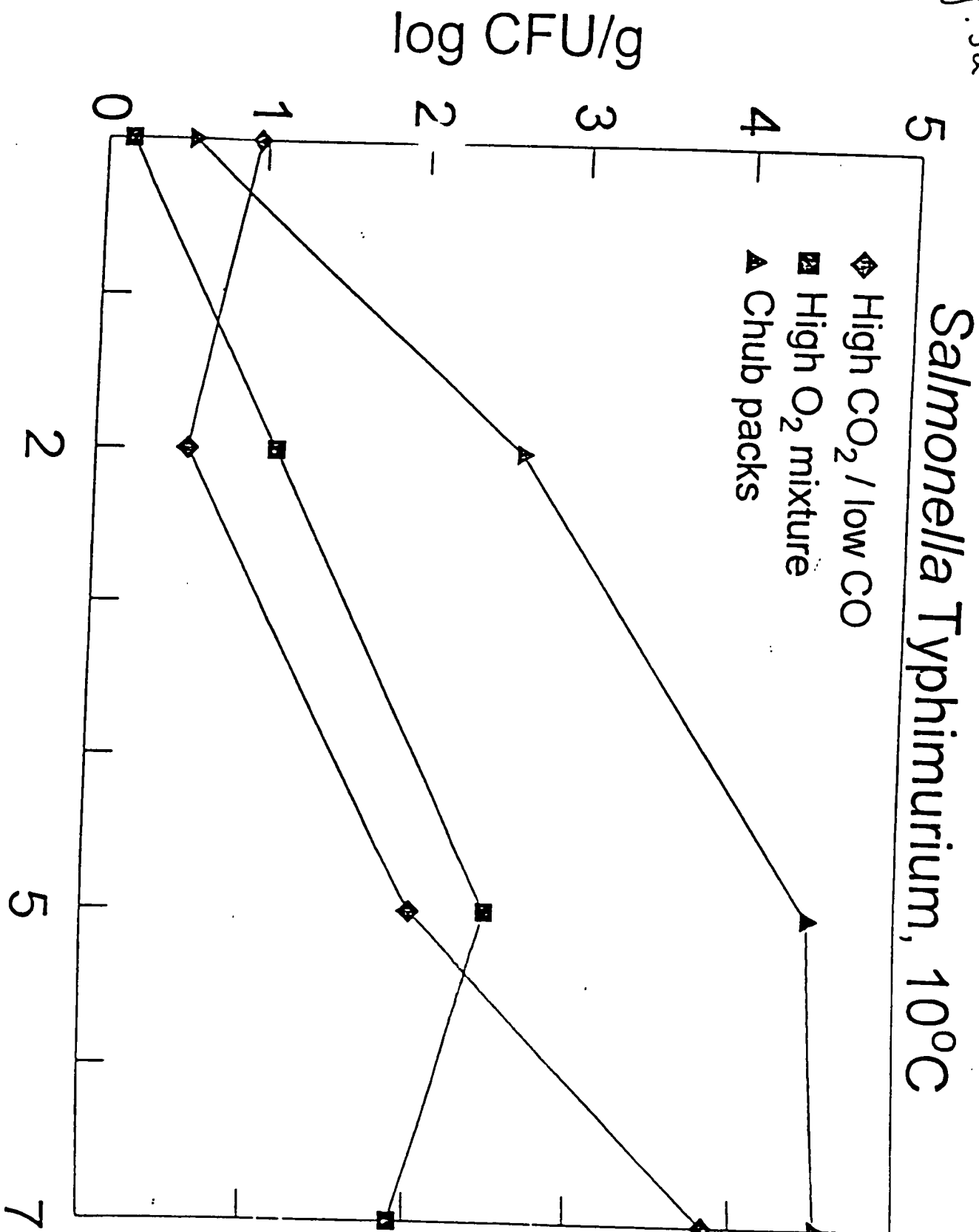
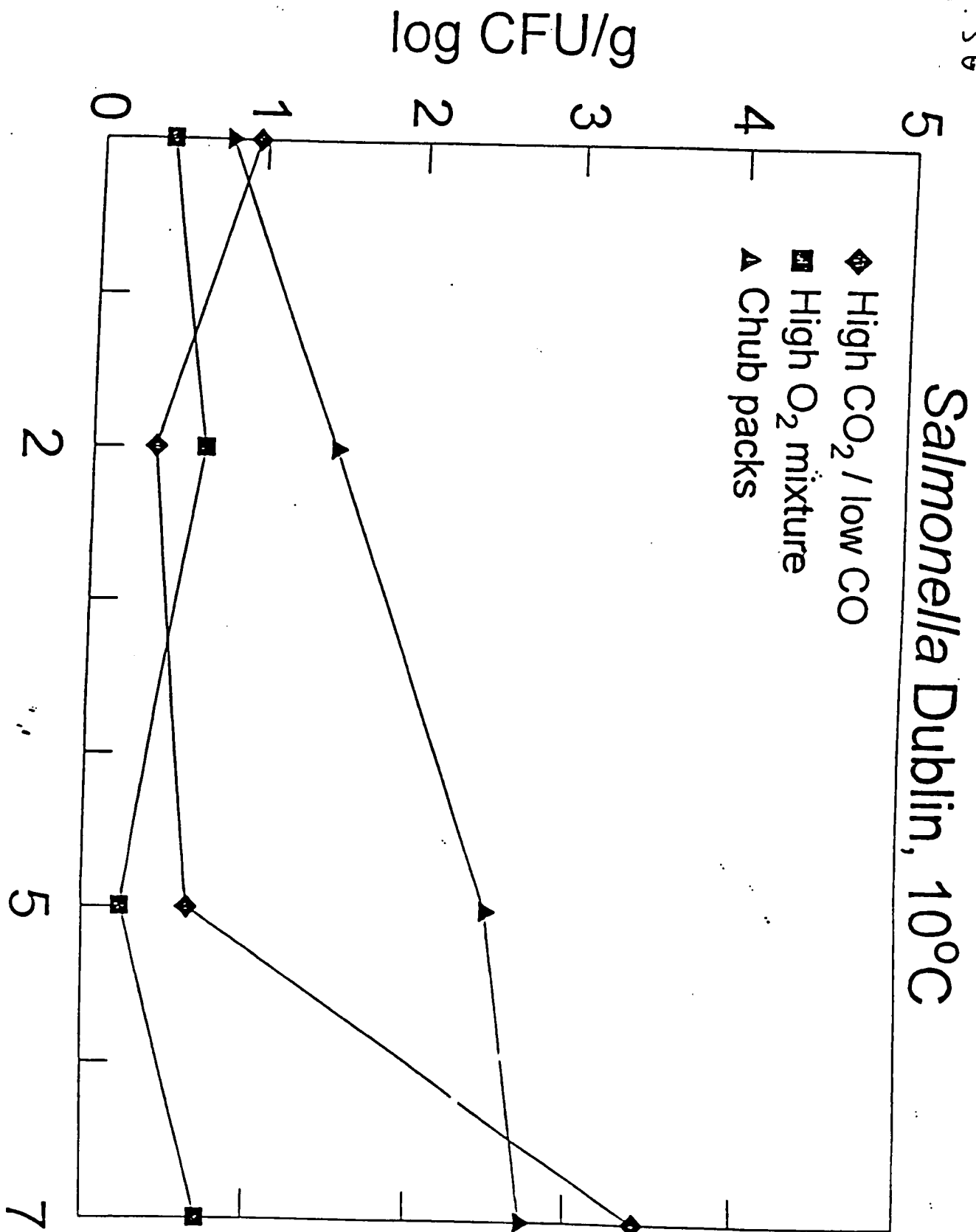
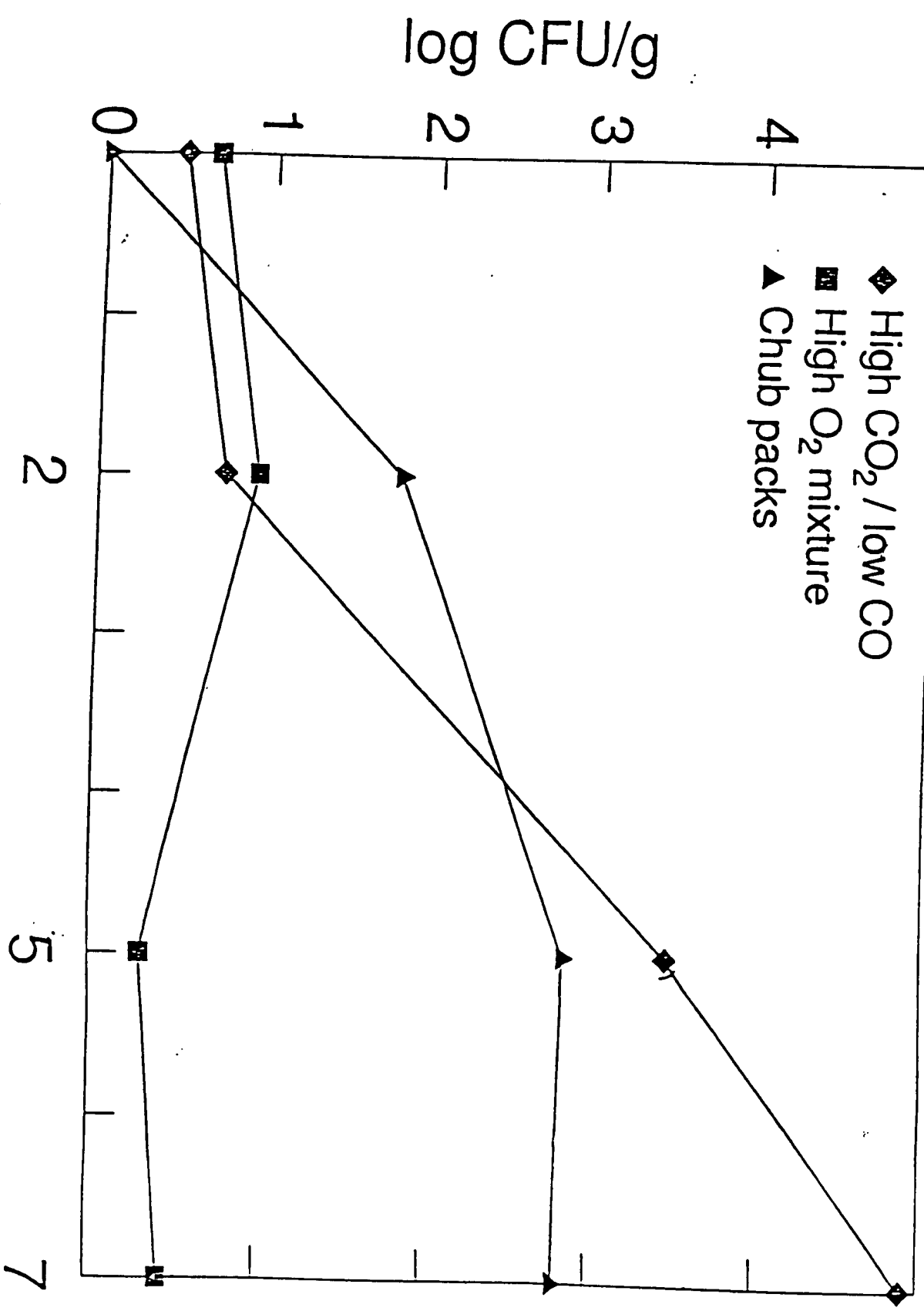


Fig. 5a

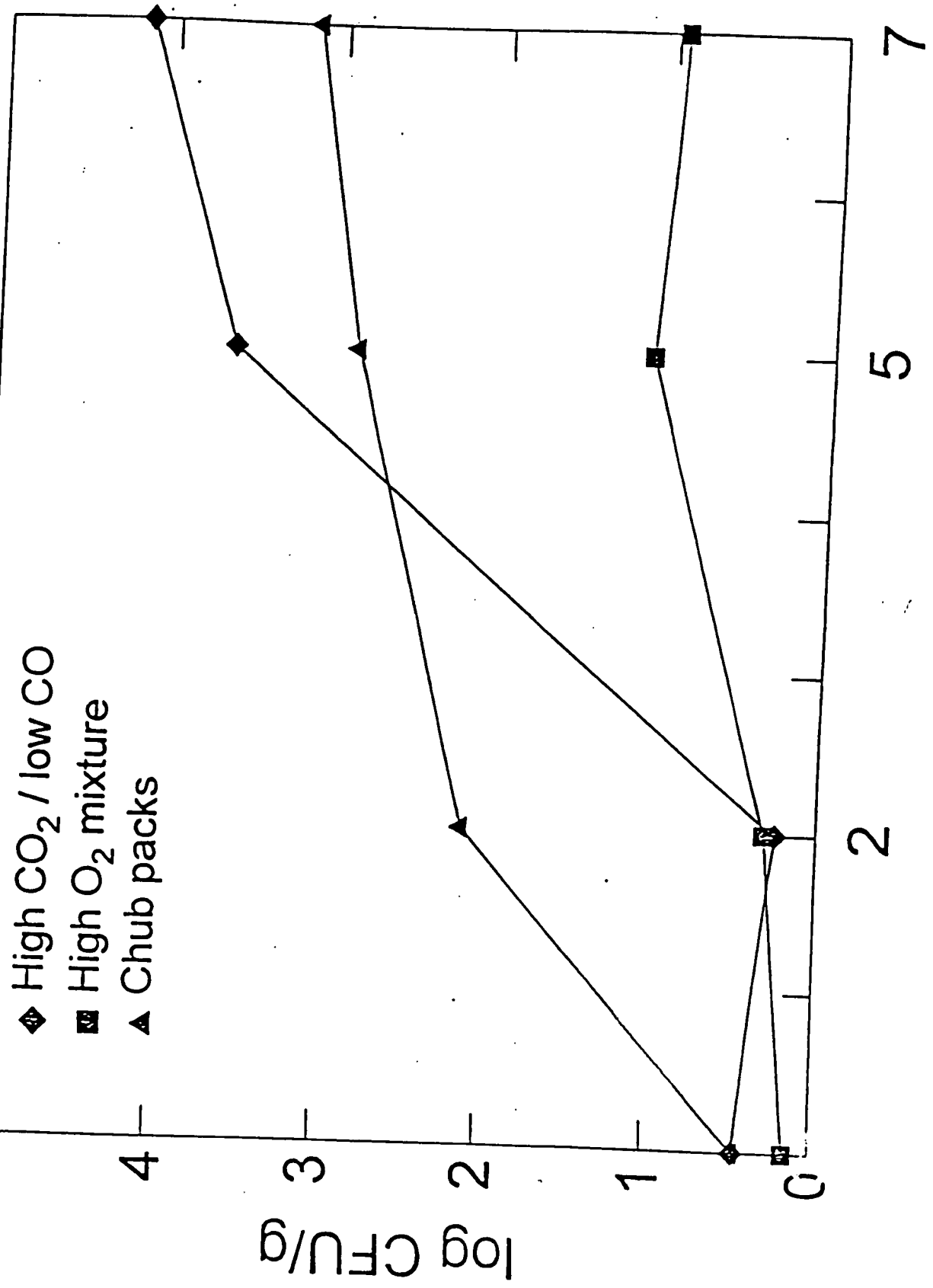


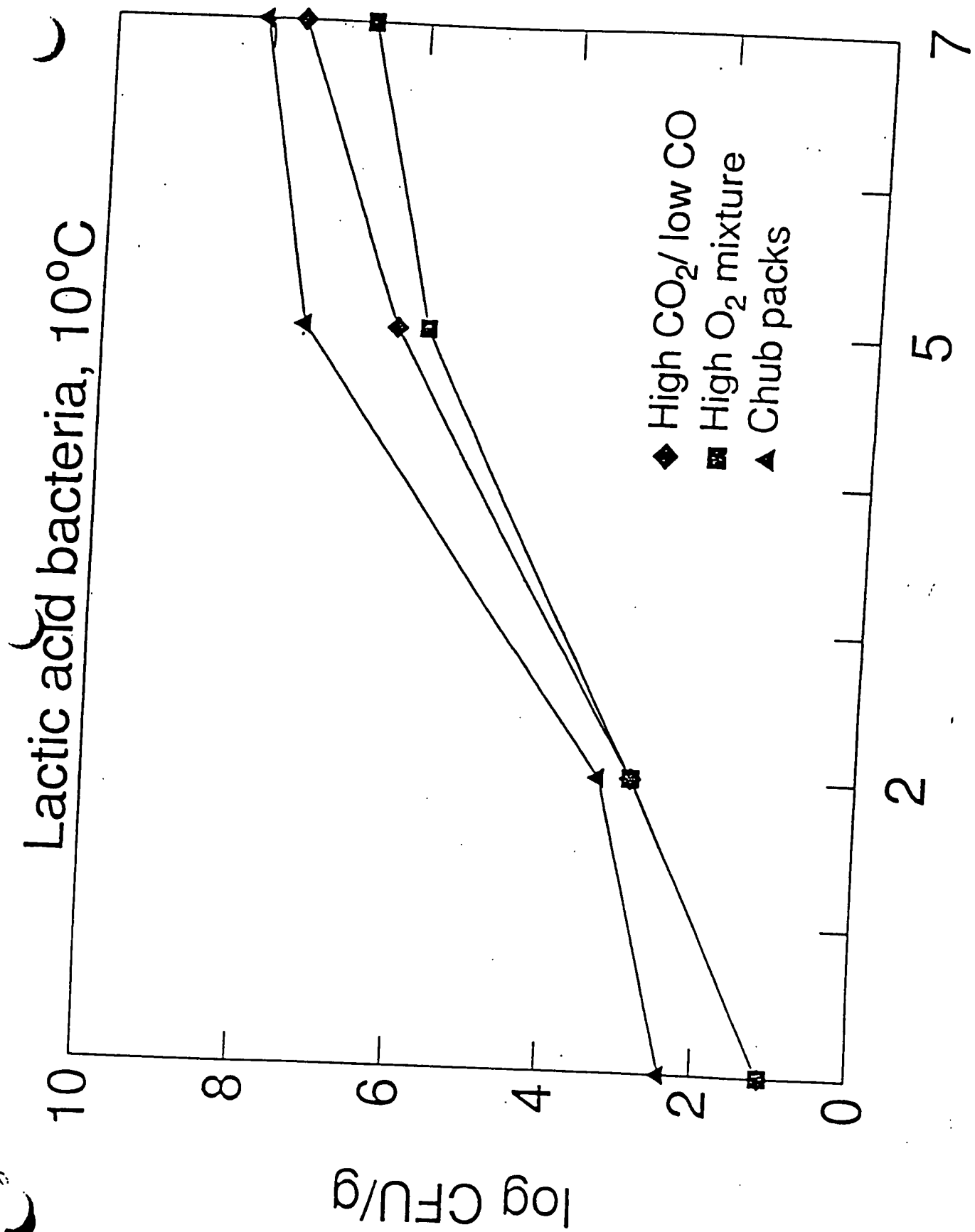


Salmonella Enteritidis, 10°C



Salmonella enterica 61:K:1,5,(7)





(2)

FOOD MICROBIOLOGY AND FOOD SAFETY INTO THE NEXT MILLENNIUM

Proceedings of the
Seventeenth International Conference of the
International Committee on
Food Microbiology and Hygiene (ICFMH)
Veldhoven, The Netherlands, 13-17 September 1999

Edited by
A.C.J. Tuijtelars¹
R.A. Samson²
F.M. Rombouts³
S. Notermans¹

¹TNO Nutrition and Food Research Institute, P.O.Box 370, 3700 AJ Zeist, The Netherlands
²Centraal Bureau voor Schimmelcultures, P.O.Box 273, 3740 AG Baarn, The Netherlands
³Wageningen University and Research Centre, Bomenweg 2, 6703 HD Wageningen,
The Netherlands

misinterpreted in the absence of storage trials or in trials of short duration such as 14 days. If *Brevibacterium thermosphacta* is a problem organism in a processing facility or on a particular type of meat, lysozyme, Chitinase or mixtures of the two could be used to control its growth during refrigerated anoxic storage.

PACKAGING OF GROUND BEEF IN AN ATMOSPHERE WITH LOW CARBON MONOXIDE AND HIGH CARBON DIOXIDE RESTRAINS GROWTH OF *ESCHERICHIA COLI* O157:H7, *LISTERIA MONOCYTOGENES*, *YERSINIA ENTEROCOLITICA* AND *SALMONELLA DIARIZONAE*

Nissen H., Alvseike O.^{1,2}, Bredholt S., Holck A. and Nesbakken T.²
MATFORSK, Norwegian Food Research Institute, Osloveg. 1, N-1430 Ås, Norway,
¹ Norwegian College of Veterinary Medicine, P.O. Box 8146 Dep., 0033 Oslo
Norway, Norwegian Meat Research Centre, P.O. Box 396, 0513 Oslo, Norway
Fax: +47 6497 0333, E-mail: hilde.nissen@matforsk.no

Ground beef for retail sale is most often ready packed in modified atmosphere (MA) or in chub packs. MA packed ground beef prolongs the microbiological shelf life and also maintains an attractive red colour. For the past decade the Norwegian meat industry has been using a gas mixture of 0.3-0.5% CO, 60-70% CO₂ and 30-40% N₂ (the CO comes ready mixed in the N₂). The reason for adding CO to the gas mixture is that it will produce a long-lasting cherry-red colour of the meat (Sørheim et al 1999). The most commonly used gas mixture for retail-ready meat in other European countries is 70% O₂/30% CO₂ (Gill 1996). The high oxygen concentration is needed to keep the red colour of the meat. It is therefore only possible to obtain half the CO₂ concentration used in the CO gas mixture. The microbiological shelf life will be longer than in air, but less than in the CO gas mixture (Sørheim et al 1999).

The inclusion of CO is controversial because the stable cherry-red colour can last beyond the microbiological shelf life of the meat and thus mask spoilage (Kropf 1980). However, the consumer is able to evaluate the microbiological conditions of the meat by off-odours and the shelf life based on odour is significantly longer in the CO mixture only at 4°C. Thus, extended shelf life does not necessarily imply an increased risk of growth of pathogens. In the present study we wanted to compare growth of the pathogens *Escherichia coli* O157:H7, *Listeria monocytogenes*, *Yersinia enterocolitica* and *Salmonella diarizonae*, in ground beef packed in a commercial Norwegian 0.4% CO/60% CO₂/40% N₂ mixture with growth in a high O₂ (70% O₂/30% CO₂) gas mixture and in ground beef in chub packs during storage at 4 and 10°C.

Commercial packages of ground beef (500 g) stored at 10°C were inoculated with the pathogens *Escherichia coli* O157:H7, *L. monocytogenes*, *Y. enterocolitica* and *S.*

Characterization and Preservation

S. diarizonae, and the ground beef stored at 4°C with *L. monocytogenes* and *Y. enterocolitica*. The inocula of *L. monocytogenes* and *Y. enterocolitica* were cocktails of 3 stationary-phase, rifampicin-resistant strains, the inoculum of *E. coli* O157:H7 was one non-toxic nalidixic/streptomycin resistant strain and that of *S. diarizonae* was a cocktail of 3 strains that were not made antibiotic resistant (plated on selective media for *Salmonella* spp.). Controls of ground beef without inoculated pathogens were stored at both temperatures.

After 5 days storage at 10°C the ground beef packed in the CO mixture had an acceptable smell while beef packed in the high O₂ mixture and the chub packs had a slight off-odour. After 8 days storage there was a strong off-odour for all the treatments. At 4°C the smell was still acceptable after 14 days of storage in the CO mixture, but the high O₂ mixture and the chub packs had some off-odours. The growth of pathogens was restrained in all samples that had been packed in the gas mixture containing CO. Thus, growth of *Y. enterocolitica* was nearly totally inhibited both at 4 and 10°C, while the number in the samples packed in the high O₂ mixture increased from about 5x10² bacteria per g at day 0 to about 10⁴ at day 5 at 4°C and to 10⁵ at 10°C. The number in the chub packs were even higher. *L. monocytogenes* showed very little growth at 4°C in all of the treatments. At 10°C there was slow growth (from about 5x10³ bacteria/g to about 10⁴ at day 5) in the CO mixture while the number in the high O₂ mixture and the chub packs were about 10 times higher. Growth of *E. coli* O157:H7 at 10°C storage was slow both in the CO-mixture and the high O₂ mixture. Growth in the chub packs was higher reaching 10⁵ bacteria/g on day 5. The growth of *S. diarizonae* followed the same pattern as *E. coli* O157:H7.

Ground beef is a high-risk product because pathogens may be mixed into the product which may not be properly heated before being eaten. The present study shows that the reduced background flora of beef packed in the CO mixture did not result in increased growth of the pathogens. This was probably due to the high concentration of CO₂ in this mixture which particularly inhibits Gram negative bacteria. The O₂ content in the CO mixture was virtually zero throughout storage at both temperatures. At 10°C the O₂ content in the high O₂ gas mixture decreased from 70% to about 35% after 8 days, probably due to aerobic bacterial metabolism. The chub packs had air-permeable casing which probably was the cause of the high bacterial growth in these packs.

The conclusion of the present study is that for the conditions studied, the risk of growth of the pathogens *Y. enterocolitica*, *L. monocytogenes*, *E. coli* O157:H7 and *S. diarizonae* in ground beef stored in CO gas mixture is the same as or less than in the ground beef stored in high O₂ or under vacuum (chub packs).

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English summary

3

CONSUMER PURCHASE PROBABILITY OF BEEF AND PORK PACKAGED IN DIFFERENT ATMOSPHERES

Ragnhild Solheim, MATFORSK, Norwegian Food Research Institute, Oslovn. 1, 1430 Ås, Norway

Ground beef, beef loin steaks and pork chops were packaged in modified atmospheres of 0.4% CO/ 60% CO₂/ 40% N₂ (high CO₂/low CO mixture) and 70% O₂/ 30% CO₂ (high O₂ mixture). In addition ground beef was packaged in clipped chub packs, beef loin steaks were vacuum packaged, and pork chops were packaged in an atmosphere of 60% CO₂/ 40% N₂ with each pack containing an O₂ absorber. The purchase probability data were collected by interviewing 126 consumers usually purchasing meat and meat products. The consumers visually compared the samples within each type of meat. The consumers preferred ground beef packaged in the high CO₂/low CO mixture or the high O₂ mixture compared to ground beef packaged in clipped chub packs. Purchase probability increased when pork chops were packaged in the high CO₂/low CO mixture. Pork chops in packs containing an O₂ absorber, were rated lowest in purchase probabilities. The purchase probability for beef loin steaks was similar when packaged in the high CO₂/low CO mixture or the high O₂ mixture, and these products were preferred compared to beef loin steaks packaged in vacuum.

COMMISSIONED REPORT

MATFORSK – Norwegian Food
Research Institute

Osloveien 1, N-1430 Ås
Tel: (+47) 64 97 01 00

Report No.:

O-7224

Availability:
Confidential

Report Title:	Date:
Consumer Survey of Meat Products	July 11, 1996
Project Manager/Author:	Signature of Project Manager:
Ragnhild Solheim	[Signature]
Head of Department:	Signature of Head of Department:
Björg Egelandstal	[Signature]
Department:	Project No:
Analysis Methodology	O-7224 FBT
Commissioned by:	Commissioner's contact:
Norsk Kjøtt [Norwegian Meat]	Truls Nesbakken

000110



Summary/Abstract:

The consumers' (N=126) purchasing tendency for ground meat, pork chops and top loin of beef packaged in various ways was measured by means of a central location test. The consumers indicated their purchasing tendency on a verbal five-point scale from "Will definitely not buy" to "Will definitely buy." Moreover, the consumers estimated their buying frequency for the three different products, and provided their age and gender.

The consumer population was composed of 62 % women and 38 % men. The age distribution was roughly equivalent for both genders.

Purchasing Frequency:

The major segment (47.6 %) of the participants said that they buy ground meat two to three times a month, while 20.6 % buy ground meat once a month.

Pork chops were purchased two to three times a month by 29.4 % of the participants, while 38.1 % bought pork chops once a month. 29 % of the participants bought pork chops less frequently than once a month.

Top loin was purchased less frequently than once a month by 58.1 % of the participants in the study. 18 % said that they buy top loin once a month, and 19.4 % indicated that they buy top loin once a week.

Purchasing Tendency:

Ground meat: Ground meat packaged with CO gas received the same average score for purchasing tendency as ground meat packaged in O₂ gas, while ground meat packaged as sausage had the lowest score for purchasing tendency.

Pork Chops: Pork chops packaged in CO gas received the highest total score for purchasing tendency, while pork chops packaged in O₂ gas received the second highest total score and pork chops packaged with oxygen absorber received the lowest score.

Top Loin: Top loin packaged in CO gas and in O₂ gas had roughly the same average score for purchasing tendency, while vacuum-packaged top loin received the lowest score for purchasing tendency.

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1. Premise

The consumers' choice of meat products may be attributed to factors such as how the products are presented in their packaging. The consumers' purchasing tendency for meat products packaged in accordance with various principles was therefore measured.

2. Implementation

2.1 Materials and Survey Conditions

The survey was taken at Drøbak City [shopping center] on June 12, 1996 (Tuesday) from 10 am to 4 pm.

Ground meat, pork chops and top loin of beef packaged in accordance with various principles (Table 1) was presented to consumers who eat these types of products. The products were delivered to MATFORSK on June 8, 1996 and stored at 4°C until the survey date. Products from the same group were placed side by side on a table under lighting with a strength of approximately 2000 lux (equivalent to the light intensity of a refrigerated meat counter in a store). The products were replaced with cold stored products every three hours.

Table 1. Packaging methods for meat products tested in consumer survey

Meat Product	Sausage	Packaging Method			
		CO	O ₂	Vacuum	O ₂ w/absorber
Ground Meat	x	x	x		
Pork Chops		x	x		
Top loin of beef		x	x	x	x

x = packaging method used

2.2 Method

The products were coded with three-digit random numbers and evaluated in a systematically rotating order. The consumers indicated purchasing probability on a verbal scale and purchasing frequency for the product, and gave their age and gender (Figure 1). Following the evaluation, the verbal scale was translated into numerical values from 1 to 5, where 1=Will definitely not buy, and 5=Will definitely buy. The consumers spent between 5 and 10 minutes answering the questions.

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Dear Consumer!

We are taking a survey on the consumer opinion concerning a selection of meat products as they appear in the meat counter. We hope that you will take a minute to let the meat producers know your opinion!

What to do:

1. Please take a look at the samples of top loin.
2. Consider whether you would buy these products the way they appear, assuming they are priced the same. Evaluate the products in the order listed below and check one box for each product.

Sample marked 763

Will definitely buy ☐
 May buy ☐
 May/may not buy ☐
 May not buy ☐
 Will definitely not buy ☐

Sample marked 288

Will definitely buy ☐
 May buy ☐
 May/may not buy ☐
 May not buy ☐
 Will definitely not buy ☐

Sample marked 911

Will definitely buy ☐
 May buy ☐
 May/may not buy ☐
 May not buy ☐
 Will definitely not buy ☐

In closing please answer the questions below about your age, gender and how often you buy top loin of beef.

My age (check one):

18-25 years old ☐
 26-35 years old ☐
 36-45 years old ☐
 46-55 years old ☐
 56-65 years old ☐
 over 65 years old ☐

Gender (check one):

Female ☐ Male ☐

I buy top loin of beef (refrigerated):

Less than once a month ☐
 Once a month ☐
 Two to three times a month ☐
 Once a week ☐
 More than once a week ☐

THANK YOU FOR YOUR ASSISTANCE!

Figure 1. Questionnaire for consumer survey of meat products. Corresponding forms were used for pork chops and ground meat.

000114

2.3 Consumer Population

124–126 consumers over 18 years of age participated in the survey of the three different meat products. There were a few more women than men among the participants, and there were fewer participants over 56 years of age than in the other age groups (Table 2). The age distribution among men and women was the same.

Table 2. Age and gender distribution among consumers participating in the survey.

Meat Product	No. of consumers	Gender (%)		Age (year, % distribution)					
		Women	Men	18–25	26–35	36–45	46–55	56–65	Over 65
Ground Meat	125	61.6	38.4	15.9	19.8	18.3	20.6	11.1	14.3
Pork Chops	126	61.9	38.1	15.1	18.3	19.8	19.8	11.1	15.9
Top Loin	124	63.7	36.3	14.5	21.0	16.9	21.8	12.9	12.9

3. Results

3.1 Purchasing Frequency for Meat Products

Ground meat was most frequently bought, followed by pork chops, while top loin was rarely bought by the consumers participating in this study (Table 3). There were similar purchasing frequencies in the various age and gender groups.

Table 3. Purchasing Frequency for Three Different Meat Products

Purchasing Frequency	Meat Product		
	Ground Meat (N=125)	Pork Chops (N=126)	Top Loin (N=124)
Less than once a month	11.1	29.4	58.1
Once a month	20.6	38.1	18.5
Two to three times a month	47.6	29.4	19.4
Once a week	15.1	3.2	3.2
More than once a week	5.6	0.0	0.8

3.2 Purchasing Tendency for Meat Products

A detailed overview of the purchasing tendency is provided in Attachment 1.

All differences described in the following were significant to a degree of 95 %.

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Ground Meat

Ground meat packaged as sausage was least appreciated by the consumers (see Figure 2). Ground meat packaged in CO gas and ground meat packaged in O₂ gas received the same average score for purchasing tendency. This result occurred regardless of consumer age and gender.

Comments from the consumers:

The wrapping film used to pack meat as sausage hides the contents.

[Above bar chart]

Purchasing Tendency for Ground Meat

[Beside bar chart] Average score (N=126) [Bar chart]

[Below bar chart]

As Sausage

CO gas
Packaging Method

O₂

Figure 2. Purchasing tendency for meat products. 1=Will definitely not buy, and 5=Will definitely buy the product as presented in the survey.

000116



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Pork Chops

Pork chops packaged in CO gas received the highest average score for purchasing tendency, pork chops packaged in O₂ gas received the second highest score and pork chops packaged with oxygen absorber received the lowest average score (see Figure 3). This result occurred regardless of consumer age and gender.

Comments from consumers:

The pork chops packaged with absorber look gray/brown – are they old? expired?

Sample packaged with CO and O₂: nitrite added?

[Above bar chart]

Purchasing Tendency for Ground Meat

[Beside bar chart]

Average score (N=126)

[Bar chart]

[Below bar chart]

O₂ absorber

CO gas
Packaging Method

O₂

Figure 3. Purchasing tendency for meat products. 1=Will definitely not buy, and 5=Will definitely buy the product as presented in the survey.

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Top Loin

Top loin packaged in CO gas and in O₂ gas received approximately the same average score for purchasing tendency (see Figure 4). Vacuum packaged top loin received a lower score for purchasing tendency than the two aforementioned samples. Men indicated roughly equivalent purchasing tendencies for the three varieties, while women indicated the highest purchasing tendency for top loin packaged in CO gas, followed by top loin packaged in O₂ gas, and the lowest purchasing tendency for vacuum packaged top loin.

Comments from consumers:

Sample packaged in CO and O₂: nitrite added?

Sample packaged in CO: "artificial" sides.

Vacuum packaged sample: looks as if it has been squeezed.

[Above bar chart]

Purchasing Tendency for Top Loin

[Beside bar chart] Average score (N=125) [Bar chart]

[Below bar chart]

Vacuum

CO gas
Packaging Method

O₂

Figure 4. Purchasing tendency for meat products. 1=Will definitely not buy and 5=Will definitely buy the product as presented in the survey.

000118

4. Conclusion

Ground Meat: Ground meat packaged in CO gas and ground meat packaged in O₂ gas received the same average score for purchasing tendency, while ground meat packaged as sausage received the lowest score for purchasing tendency.

Pork Chops: Pork chops packaged in CO gas received the highest score for purchasing tendency, pork chops packaged in O₂ gas received the second highest total score, while pork chops packaged with oxygen absorber received the lowest score.

Top Loin: Top loin packaged in CO gas and in O₂ gas received approximately the same average score for purchasing tendency, and vacuum packaged top loin received the lowest score for purchasing tendency.

Comments

This type of survey does not have representative sampling of consumers in terms of the population as a whole or a specific population segment. The make up of the survey represents a model for a purchasing situation. These circumstances must be taken into account when interpreting the results.

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ATTACHMENT 1

Overview of Results from Consumer Study of Meat Products

	<i>Average</i>	<i>Standard Deviation</i>	<i>Median</i>
<i>Ground Meat (N=126)</i>			
As sausage	2.5	1.5	2
CO gas	3.7	1.4	4
O ₂	3.7	1.4	4
<i>Pork Chops (N=126)</i>			
O ₂ absorber	1.9	1.3	1
CO gas	4.6	0.7	5
O ₂	3.6	1.4	4
<i>Top Loin (N=125)</i>			
Vacuum	2.9	1.6	3
CO gas	4.2	1.1	4.5
O ₂	4.0	1.1	4

000120

Prosjektmedarbeidere:

Ansvarlig for *opplegg og rapport*: Ragnhild Solheim

Ansvarlig for *prøvemottak og lyssetting*: Oddvin Sørheim

Ansvarlig for *praktisk gjennomføring*: Inger Johanne Fjøsne

Øvrige deltakere i gjennomføring:

Asgeir Nilsen
Janne Haugdahl
Vibeke Høst
Oddvin Sørheim

Ansvarlig for *statistisk bearbeiding*: Per Lea

English summary

DISCOLORATION OF MEAT AS AN INDICATOR OF LEAKAGES IN PACKAGES CONTAINING A CO GAS MIXTURE

Oddvin Sørheim, MATFORSK, Norwegian Food Research Institute, Oslovn. 1, 1430 Ås, Norway

The aim of the experiment was to study discoloration of meat packaged in a gas mixture of 60 % CO₂/40 % N₂/ 0.4 % CO with different concentrations of residual O₂ added. Tests were performed on ground beef with 1 % NaCl, aged beef loin steaks and pork chops. Leakages were simulated by injecting different amounts of air with a syringe into the packages after two days storage. Discoloration of the meat was measured as reduction in a* (redness) values and evaluated visually. Ground beef had a low tolerance level of residual O₂ because it was discoloured in atmospheres containing more than 1 % O₂. Beef loin steaks and pork chops were slightly discoloured in more than 2 and 5 % O₂, respectively. The results suggest that discoloration can be an indicator of leakages for ground beef, but not for beef loin steaks and pork chops.

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**MATFORSK – Norwegian Food
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Osloveien 1, N-1430 Ås
Tel: (+47) 64 97 01 00

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Project Manager/Author: Oddvin Sorheim	Signature of Project Manager: [Signature]
Head of Department: Astrid Nilsson	Signature of Head of Department: [Signature]
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Summary/Abstract:

Tests were carried out to find the tolerance limits for residual O₂ for discoloration of meat packaged in CO gas mixture with simulated leak. Various concentrations of air were added to the packages of ground meat, top loin and pork chops with a mixture of 60% CO₂ / 40% N₂ / 0.4% CO two days after packaging. Ground meat packaged in gas containing more than 1% O₂ was clearly discolored, while top loin and pork chops, respectively packaged in gas containing more than 2 and 5% O₂ showed only minor discoloration. The results indicate that discoloration can serve as an indicator of leakage for ground meat, but not for top loin and pork chops.

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Meat Discoloration as an Indicator of Leakage in Packages with CO Gas Mixtures

Purpose

The purpose of the survey was to find the tolerance limits for residual O_2 with regard to discoloration of ground meat, top loin and pork chops packaged in a CO gas mixture.

Implementation of the Study

Samples of ground meat with 1% NaCl (20 pieces), tenderized top loin of beef (18 pieces) and pork chops (14 pieces) were gas packed on a Ilapak Delta 2000 machine (Ilapak, Switzerland) on a tray in BDF 550 shrink film (Cryovac). The gas mixture consisted of 60% CO_2 / 40% N_2 / 0.4% CO. The samples were stored out of light at 4°C. After two days of storage, air was added to the packages to increase the O_2 content, i.e. a simulated leak. This was done by sucking out the gas in the package and replacing it with air by means of a syringe and a septa. 0-2.0% O_2 was added to the ground meat, 0-3.2% O_2 was added to the top loin, and 0-13.9% O_2 was added to the pork chops, in all cases with a spectrum of O_2 concentration in their respective ranges. After the replacement of gases, the concentrations of O_2 and CO_2 were measured by means of two Toray instruments, type LC 700F and PG-100 (Toray Eng., Japan). The remaining storage time before unwrapping was 2 days for ground meat and 5 days for both top loin and pork chops.

Upon unwrapping, the O_2 and CO_2 concentrations in the packages were once again measured. Two judges then visually judged the color of the unopened packages according to a scale (1=fresh meat red, 2=dark red, 3=somewhat discolored, 4=moderately discolored, 5=extremely discolored). The packages were then opened, and the color was measured with a Minolta Chroma Meter CR-300 (Minolta Camera Co., Japan) directly on the surface of the meat within 1 minute of opening. The instrument had light source D_{65} at 8 mm aperture, and the color was measured in CIE (1976) L^* (luminosity), a^* (redness) and b^* (yellowness). Lastly, the pH was measured directly in the meat with a Ingold Xerolyt electrode (Mettler-Toledo A.G., Switzerland).

Results and Discussion

The correlation between discoloration upon unwrapping and O_2 concentration when replacing the gas proved to best be expressed by an a^* value (redness) and visual color evaluation. Attached are a plot of the a^* and O_2 concentration for ground meat, top loin and pork chops; see figures 1, 2, and 3. The correlation coefficients for the three products were calculated to -0.71, -0.33, and -0.51. The relatively low coefficients are partly due to the large spread of the measured values and partly because the correlation between a^* and O_2 does not appear to be linear.

For ground meat we found a reduction of approximately 4-5 a^* values from 0 to 1% O_2 . A reduction of a^* to this degree is readily apparent. Samples stored in 1% or higher levels of O_2 had a score of between 3 and 4 on the color scale, i.e. slight to moderate discoloration. The results indicate that the tolerance limit for discoloration of ground meat in CO mixture is approximately 1% O_2 .

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For top loin, on the other hand, a smaller decrease in a^* and a weak correlation between a^* and O_2 concentration were found. However, there seemed to be a reduction of about 2 a^* values between 0 and 2% O_2 , with a color scale score of about 3 on samples stored in over 2% O_2 . This indicates a weak discoloration with a tolerance limit of approximately 2 % O_2 for top loin.

The color of pork chops proved to be only slightly affected by the O_2 concentration in the packaging gas, even when up to 2/3 of the gas was replaced with air. We found a reduction of 1 to 1.5 a^* values between 0 and 5% O_2 in the package gas, but this barely registered as discoloration with a score of 2-3 on the color scale.

The pH values at the end of the leak test were on average 5.59, 5.62 and 5.42 for ground meat, top loin and pork chops respectively.

Between the start and the end of the leak tests, we measured a reduction in the O_2 concentrations of 80, 40 and 30 % for ground meat, top loin and pork chops respectively. This reduction can be due to meat respiration or consumption of O_2 by bacteria. Ground meat has a high consumption of O_2 due to a large surface area exposed to surrounding gas and frequently higher bacteria counts than whole meat.

The significance of residual O_2 in package gas with regard to discoloration and microbiological storage life has been discussed previously in the report "Fresh Meat in Consumer Packaging - an Evaluation of Various Packaging Methods and Their Effect on Meat Quality." For storage in gas containing CO_2 and/or N_2 without the presence of CO , tolerance limits for discoloration have been found to be below 0.1 and 0.5% O_2 for beef and pork respectively. Tests on pork has shown that the microbiological storage life was reduced when the package gas contained more than 2-4% O_2 .

The ground meat containing 1% sodium that was tested in this survey, had obvious discoloration when the CO mixture contained at least 1% O_2 . Sodium functions as a pro-oxidant, and will usually intensify or accelerate the discoloration of the meat. It is therefore likely that consumers will react on the color of ground meat when small leaks in the packaging exist. For top loin and pork chops, however, there is little likelihood that the minor discoloration occurring at above 2 and 5% O_2 will serve as an indicator of leakage to the regular consumer. The lighting in store refrigerating counters will often conceal minor color nuances. All in all, these results show that CO has a strong bond to the myoglobine in whole, unsalted meat, which prevents the carbon myoglobine from being destabilized by O_2 in the gas. Hence, discoloration is not a good indicator with regard to alerting consumers of leaks and risk of increased bacterial growth in meat such as top loin and pork chops.

Thanks

We are grateful to Frank Lundby for his valuable technical assistance.

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[Left-hand side of graph] a^*

[Plot]

[Below graph]

% Oxygen

Figure 1 The correlation between a^ (redness) and O_2 concentration for ground meat packaged in a mixture of 60% CO_2 / 40% N_2 / 0.4% CO after two days storage at 4°C. $n=20$, $r=-0.71$.*

[Left-hand side of graph] a^*

[Plot]

[Below graph]

% Oxygen

Figure 2 The correlation between a^ (redness) and O_2 concentration for top loin packaged in a mixture of 60% CO_2 / 40% N_2 / 0.4% CO after five days storage at 4°C. $n=18$, $r=-0.33$.*

[Left-hand side of graph] a^*

[Plot]

[Below graph]

% Oxygen

Figure 3 The correlation between a^ (redness) and O_2 concentration for pork chops packaged in a mixture of 60% CO_2 / 40% N_2 / 0.4% CO after five days storage at 4°C. $n=14$, $r=-0.51$.*

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Review

Retail meat can be packaged in gas mixtures containing 60–70% carbon dioxide (CO_2), 30–40% nitrogen (N_2) and <0.5% carbon monoxide (CO). This gas mixture with CO provides a unique combination of a long microbiological shelf life and a stable, cherry red colour of the meat. The shelf life of meat packaged in the CO mixture is longer than that of meat packaged in the commonly used atmospheres with high oxygen (O_2), that is, approximately 70% O_2 and 30% CO_2 . The consumption of meat that has been packaged in a CO mixture will result in only negligible levels of carboxyhaemoglobin in the blood. It is highly improbable that the use of CO in the packaging of meat will present a toxic threat to consumers.

Modified-atmosphere packaging (MAP) is gaining increasing application in modern food distribution. Meat intended for retail sale can either be wrapped in vapour-tight, oxygen-permeable films or packaged in gas-tight films with a modified atmosphere (MA). The main purposes of the MAP of meat are twofold: to ensure the microbiological shelf life and the attractive red colour of the product. Consumers frequently interpret the colour of meat on retail display as an indicator of wholesomeness¹.

CO is a colourless, odourless and tasteless gas. It is produced mainly through incomplete combustion of carbon-containing materials. Natural background levels of CO are 0.01–0.9 mg/m³ (Ref. 2). In urban areas, 8-h mean concentrations of CO (i.e. mean CO concentrations are measured for each possible 8-h interval during a 24-h period, then averaged) are generally <20 mg/m³; however, maximum 8-h concentrations (i.e. the maximum mean concentration found during any one 8-h period) of up to 60 mg/m³ have been recorded². By far the most common cause of elevated CO concentrations in the blood is tobacco smoking³.

A challenge in the MAP of retail meat is the stabilization of the red colour of the product. The positive effect of CO on meat colour was known and patented over 100 years ago⁴. Despite this knowledge, CO has to date been applied commercially only to a limited extent in the MAP of meat. During the past 10 years, the Norwegian meat industry has been using a gas mixture of 60–70% CO_2 , 30–40% N_2 and 0.3–0.4% CO for the packaging of fresh retail meat, namely beef, pork and lamb. This gas mixture with CO maintains a stable, cherry red colour combined with a long microbiological shelf life

Technological, hygienic and toxicological aspects of carbon monoxide used in modified-atmosphere packaging of meat

Oddvin Sørheim, Tore Aune and Truls Nesbakken

of the meat. The market share of retail meat packaged in this CO mixture in Norway is estimated at 50–60% (Dag Hallan, pers. commun.). In addition, some meat is also initially bulk-packaged in the CO mixture, and thereafter repackaged on trays with O_2 -permeable films in retail outlets. In other European countries not using such CO mixtures, market shares of retail meat packaged in atmospheres with a high O_2 concentration, with considerably shorter shelf lives, have been reported to be only 10–40%^{5,6}.

In this article, we have evaluated the toxicological aspects of CO, and its mode of action and application in the MAP of meat.

Technological and hygienic aspects of CO as a packaging gas for meat Gases for MAP

The most commonly used gases for the MAP of meat are CO_2 , N_2 and O_2 , although other gases, including CO, nitrous oxide, argon and ozone, have been tried to a limited extent⁷. CO_2 inhibits the growth of many microorganisms, but it has no effect *per se* on the colour of meat⁸. CO_2 is absorbed in meat and fat tissue at a ratio of ~1 litre of gas per kg of tissue⁹. N_2 affects neither the microbiology nor the colour of the meat, but prevents packages from collapsing, because it is not absorbed by the product. O_2 supports the growth of aerobic microorganisms; thus, removal of O_2 from the MA will extend the microbiological shelf life. High O_2 concentrations cause meat to have a temporary bright red colour; oxygen binds to the muscle pigment myoglobin, forming oxymyoglobin, which is gradually oxidized to grey-green-brown metmyoglobin⁸ (Fig. 1). Gases for the packaging of meat are seldom used alone, but in mixtures, which vary according to the application. Examples of different gas mixtures for the MAP of meat

Oddvin Sørheim is at MATEFORSK – Norwegian Food Research Institute, Osloveien 1, N-1430 Ås, Norway (fax: +47-64-970333; e-mail: oddvin.sorheim@matforsk.no). Tore Aune is at the Department of Pharmacology, Microbiology and Food Hygiene, Norwegian College of Veterinary Medicine, PO Box 2145 Dep., N-0033 Oslo, Norway. Truls Nesbakken is at the Norwegian Meat Cooperative, Department of Research and Development, PO Box 360 Økern, N-0513 Oslo, Norway; and at The Royal Veterinary and Agricultural University, Department of Veterinary Microbiology, Bülowsvej 13, DK-1870 Frederiksberg C, Denmark.

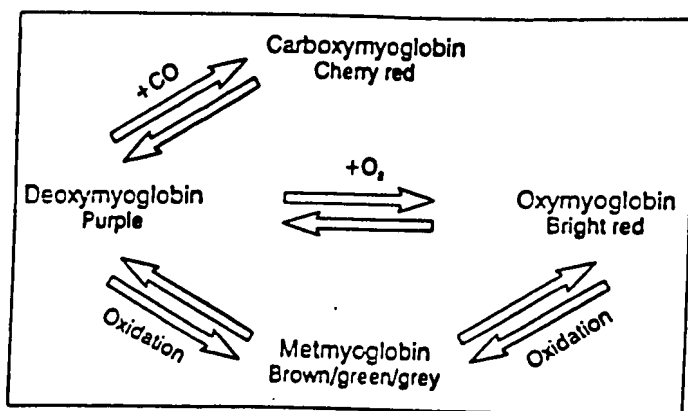


Fig. 1
Myoglobin forms and colour of meat

CO and colour

The main function of low levels of CO in MAs is to give meat a stable, cherry red colour, as a result of strong binding of CO to myoglobin and the formation of carboxymyoglobin⁹ (Fig. 1). Although a substantial increase in the shelf life of meat can be obtained by using various MAs, it is often limited by discolouration due to the oxidation of myoglobin to metmyoglobin. This discolouration can be prevented by the inclusion of a low level of CO in the gas mixture.

Carboxymyoglobin is more resistant to oxidation than oxy-myoglobin, owing to the stronger binding of CO to the iron-porphyrin site on the myoglobin molecule¹⁰. CO at concentrations of 1–5% increased the reduction of metmyoglobin, even in the presence of air¹¹.

Examples of different gas mixtures that contain CO for the packaging of meat are given in Table 1. A mixture of 2% CO and 98% air was very effective in stabilizing the colour of beef for 15 d, compared with 5 d in air alone¹². Ground beef patties stored in an atmosphere

of 1% CO/50% CO₂/49% air retained a stable, red colour for at least 6 d, whereas the colour of samples stored in air was stable for only 3 d¹³.

The colour of beef was analysed during storage in MAs containing N₂ with 0.5–10% CO. Levels of CO >0.5% resulted in a stable, red colour for >30 d, whereas discolouration occurred after 5 d storage in control samples packaged in air¹⁴. In addition, samples of this beef were exposed to pure CO for 2–16 h before packaging in air. The colour stability of the CO-treated samples was no greater than that of untreated samples. However, in other experiments the exposure of beef to CO before vacuum packaging increased its redness and colour acceptability during subsequent chilled or frozen storage^{9,17}. Beef loin roasts stored in 1% CO/51% CO₂/30% O₂/18% N₂ were shown to have lower levels of metmyoglobin on their surface than vacuum-packaged roasts. After a further 4 d on retail display, steaks from the roasts underwent less discolouration if they had previously been stored in the CO mixture¹⁵. Ground beef and beef loin steaks packaged in 1% CO/50% CO₂/25% N₂/24% O₂ or 1% CO/20% CO₂/9% N₂/70% O₂ retained a stable colour for 29 d¹⁶. Similarly, beef loin steaks packaged in 0.4% CO/60% CO₂/40% N₂ maintained a stable, cherry red colour for up to 22 d¹⁸. Experiments with beef and higher levels of CO, that is, 2% CO/20% CO₂/78% N₂, resulted in meat that had a stable colour; however, its colour was characterized as 'too artificial' by a sensory panel¹⁴.

Based on the cited literature, the presence of 0.4–1.0% CO in MAs used for the packaging of meat seems sufficient to produce a stable, cherry red colour.

Cooked, cured meat products can also benefit from storage in MAs containing CO. Packaging in 1% CO/99% N₂ stabilized the colour of sliced bologna, indicating binding between CO and denatured myoglobin¹⁹.

Under certain circumstances, an undesirable pink or red colour can arise in cooked white meat, such as poultry, and cooked meat products without added nitrite²⁰. Such colour problems can sometimes be linked with exposure to CO, which results in similar colours occurring after the use of MAs with CO. For example, roasted turkey was noted to be pink; this was probably due to the presence of CO and nitric oxide in the combustion gases in the oven. The pink colour did not occur when the turkeys were roasted in complete isolation from the oven gases²¹. Combustion engines produce various gases, including CO, which can affect live poultry during transportation to the abattoir. Meat from chickens that were exposed to exhaust fumes immediately before slaughter developed an undesired red colour on cooking²².

CO and microbiology

Generally, the purpose of most of the experiments investigating the use of CO as a small component of MAs for meat has been to study its effect on colour stability, and more seldom its microbiological aspects. The growth of psychrotrophic bacteria on beef stored in MAs containing 0.5–10% CO in N₂ was lower, relative to controls, resulting in a longer shelf life²³.

Table 1. Applications of carbon monoxide (CO) in the modified-atmosphere packaging of meat

Gas combinations (%)					Refs
CO	CO ₂	N ₂	O ₂	Air	
2				98	12
1	50			49	13
0.5–10		90–99.5			14
1	51	18	30		15
1	50	25	24		16
1	20	9	70		16
2	20	78			6
1–5				95–99	10
100 ^a					9, 14, 17
0.4	60	40			18
0.3–0.4	60–70	30–40			b

^aExposure before packaging

^bData supplied by Norwegian meat plants

temperatures in the range of 0–10°C¹⁴. For example, beef packaged in a MA of 1% CO/99% N₂ had an odour shelf life of 24 d, compared with 18 d in 100% N₂, and 7 d in air at 5°C. However, in another experiment with a MA of 20% CO₂/70% O₂/9% N₂, the addition of 1% CO had no effect on the microbiological growth on ground beef and beef steaks¹⁶. The presence of bacteriostatic CO₂ in the latter experiment apparently reduced the importance of the effect of CO on the shelf life of the meat. The odour shelf life of steaks of beef loins stored in 0.4% CO/60%

CO/40% N₂ was 4 d longer than that of steaks stored in 70% O₂/30% CO₂ at 4°C¹⁸. Beef steaks that were exposed to pure CO before vacuum packaging had an extended shelf life compared with untreated controls. The total aerobic plate, lactic acid bacteria and psychrotropic counts of CO-treated steaks were 1–2 log cycles lower than those of controls after 8 weeks storage at 4°C¹⁷. In a study using pure bacterial cultures, the presence of CO at a concentration of 5–30% in air had no effect on the growth of *Pseudomonas aeruginosa*, inhibited the growth rate of *Escherichia coli* (in proportion to the concentration of CO), increased the lag phase of *Achromobacter* and inhibited the growth rate of *Pseudomonas fluorescens*²¹.

Toxicological aspects of CO

Health effects of CO

CO binds to the iron atom of haemoglobin in red blood cells, forming carboxyhaemoglobin (COHb). The affinity of haemoglobin for CO is ~240 times higher than its affinity for O₂. CO also binds to myoglobin, cytochromes and some enzymes, but these reactions are considered to be of less importance than the formation of COHb². The binding of CO to haemoglobin is reversible, with a half-life of ~4.5 h in individuals who are at rest.

Although CO acts primarily by interfering with O₂ transport, it also reduces the delivery of O₂ to the various tissues³. In humans, health effects are mainly manifested in the cardiovascular system, the nervous system and in the foetus.

The COHb concentration in blood, often referred to as the COHb percent (COHb%), is a function of the CO concentration in the air, the exposure time, and the level of physical activity of the individual²⁴ (see Table 2). At a COHb concentration of ~2.5%, the most sensitive individuals (patients suffering from cardiovascular diseases) display changes in cardiac function and report chest pain. At somewhat higher COHb concentrations, they experience reduced working capacity and the onset of angina pectoris on exercise^{25,26}. In healthy adults, no adverse health effects were described at CO concentrations that result in <5% COHb²⁷.

A small amount of CO is formed naturally in the human body, owing to the breakdown of haemoproteins.

Table 2. Estimate of carboxyhaemoglobin percent (COHb%) in human blood at different concentrations of carbon monoxide (CO) in the atmosphere, depending on the level of physical activity*

Exposure		COHb%		
CO concentration (mg/m ³)	Time (h)	At rest	Moderate activity	Heavy work
10	8	1.3	1.4	1.4
25	1	1.0	1.5	2.0
40	1	1.3	2.2	2.9

*Data taken from Ref. 24

Table 3. Association between different carboxyhaemoglobin (COHb) levels in blood and health effects*

COHb%	Observed health effects
≥50	Unconsciousness, lethal if not treated
≥30	Headache, nausea, vomiting, dizziness
≥10	Life threatening for heart and lung patients; headache in other individuals
≥5	Reduced maximum oxygen consumption during exercise in healthy individuals
≥5	Reduced visual perception, learning ability and fine motor performance
≥5	The foetus can be affected on carbon monoxide exposure of pregnant women
≥2.9	Angina patients endure less physical strain before experiencing attack
≥2.3	Reduced physical working capacity, especially endurance
≥2.2	Possible reduction in attention and ability to concentrate
≥2.2	Signs of local lack of oxygen and onset of chest pain in heart patients

*Data taken from Refs 25–27

Such production leads to a COHb concentration of ~0.5%. The average COHb concentration in non-smokers is 1.2–1.5%, and ~3–4% in smokers²⁷.

The absorption and excretion of CO from the body occur relatively slowly; thus, exposure to elevated CO levels over short time periods will not result in a significant increase in the COHb level in the blood. Table 3 details the various health effects observed at different COHb levels. This table confirms that exposure to CO that results in a COHb level greater than ~2% should be avoided to protect the most vulnerable individuals in the population.

In order to protect the most vulnerable in society, a Norwegian expert group on air pollution²⁷ recommended maximum CO concentrations for different exposure times that will prevent COHb levels from exceeding 1.5%, taking into consideration endogenous CO formation (Table 4).

Table 4. Estimates of carbon monoxide (CO) levels in ambient air that will result in carboxyhaemoglobin (COHb) levels of 1.5%, including endogenous CO production*

Exposure time	CO concentration in air (mg/m ³)		
	At rest	Moderate activity	Heavy work
15 min	170	80	52
30 min	86	42	29
1 h	48	24	18
8 h	11.5	9.2	9.2

*Data taken from Ref. 27

Exposure to CO on consumption of fresh meat treated with a CO gas mixture

Very little information exists in the literature on the exposure to CO following the consumption of meat that has been treated with CO gas. The inhalation of air containing CO at a level of 57 mg/m³ (the acceptable level in working environments in the USA) would provide a COHb level for a prolonged time period (hours) of at least 14 times that of the level reached temporarily on the consumption of 225 g of meat that had been packaged in CO at the saturation level for myoglobin¹⁴. In this estimate, it was assumed that the saturation of the meat myoglobin and haemoglobin was maximal and that the transfer of CO from the gastrointestinal tract to the blood was 100%. Consequently, even for such a 'worst-case' scenario, the treatment of meat with CO gas appears to contribute very little to COHb levels, relative to levels that are considered safe in the working environment. The exposure of beef to an atmosphere containing 1% CO for 3 d resulted in ~30% saturation of the meat myoglobin²¹. CO is lost from previously CO-treated meat during storage in the absence of CO, with a half-life of ~3 d. When the beef was cooked at 195°C, only 0.1 mg of CO remained per kg of meat. The loss of CO amounted to ~85%.

Comparison of CO exposure from air and the consumption of gas-treated meat

Data are very scarce, but comparisons still allow crude estimates to be made. An adult inhales ~10–20 m³ of air in 24 h (depending on their level of activity). This is equivalent to 0.42–0.84 m³/h (or 3.36–6.72 m³ in 8 h). In order to prevent a maximum COHb level in the blood of 1.5% being exceeded, the CO concentration in air for a 1-h period of moderate physical activity should not exceed 24 mg/m³, or 9.2 mg/m³ in 8 h (according to Table 4). In contrast, the consumption of meat that had been treated for 3 d in an atmosphere containing 1% CO yielded ~0.1 mg of CO per kg of meat on storage and cooking²¹. Based on these data, a comparison can be made from the two methods of exposure to CO, and is shown in Table 5.

Equilibrium between CO present in the atmosphere and the COHb concentration in blood is reached only

after a considerable period of time (depending on the concentration and level of physical activity). Even in a 'worst-case' scenario, equilibration between the CO concentration in the gastrointestinal tract and blood will take time. Furthermore, the absorption of CO from the gastrointestinal tract into the blood will in all probability be less effective than the absorption of CO from the lungs, which are composed of tissues that are designed to facilitate gas exchange between the alveoli and the blood. Consequently, it is highly probable that the consumption of one meal of CO-exposed meat per day will not result in measurable increases in the COHb level in blood.

Toxicological evaluation of the use of CO as a packaging gas for meat

Unfortunately, the European Union (EU) has not evaluated CO for use as a packaging gas for meat. However, CO₂ and nitrous oxide (N₂O) have both been approved for use for extraction purposes, and it was considered unnecessary to adopt an acceptable daily intake (ADI) value for these gases in this application²⁸.

In order to avoid possible adverse health effects in those individuals who are the most susceptible, a Norwegian expert group on air pollution recommended maximum CO concentrations in ambient air that result in COHb levels not exceeding 1.5% (including endogenous CO production)²⁷. Estimates detailed above indicate that, even assuming an improbable 100% absorption of CO from the gastrointestinal tract into the blood, the consumption of meat that has been treated with 1% CO will result in COHb levels that are negligible (approximately three orders of magnitude lower) compared with those resulting from exposure in the working environment to CO at an acceptable level. Consequently, it is highly improbable that CO exposure from meat packaged in an atmosphere containing up to 0.5% will represent a toxic threat to consumers through the formation of COHb.

Alternatives to the MAP of retail meat

Currently, the most commonly used MA for the retail packaging of meat contains O₂ at a high concentration in combination with CO₂, such as ~70% O₂/30% CO₂. The shelf life of meat in a high O₂ atmosphere in commercial practice, typically at temperatures of 6–8°C, is ~7 d, being limited both by microbiological spoilage and discoloration. Meat that is stored in a high O₂ concentration is often spoiled by bacteria such as *Brochothrix thermosphacta* and *Pseudomonads*³⁰. In MAs with a high concentration of O₂, the meat normally maintains its bright red oxymyoglobin colour for 4–7 d before the colour starts deteriorating to grey-brown, owing to the formation of metmyoglobin¹⁸. This length of time is often not considered to be sufficient to display and sell the product.

The use of MAs with a high concentration of CO₂, either alone or in combination with up to 70% N₂, would increase the microbiological shelf life of the meat compared with that of meat in a MA with a high O₂ concentration. The absence of O₂ together with the presence

of CO₂ retards microbiological growth. Unfortunately, the colour of meat packaged in MAs containing CO₂ is less satisfactory, being either purple or grey-brown due to the formation of deoxymyoglobin or metmyoglobin, respectively. The meat inevitably discolours when the O₂ concentration is low. Metmyoglobin formation can be avoided by maintain-

ing O₂ concentrations <0.01–0.1% for beef³¹ and <0.5% for pork³². These low O₂ levels, particularly for beef, are difficult to achieve in most commercial packaging operations, because a small amount of air will unavoidably be incorporated in the MAs of the packages. MAs with a high CO₂ concentration seem to be useful for retail packaging if a low concentration of CO is also included to stabilize myoglobin and the meat colour.

Vacuum packaging is commonly used for the bulk storage, transportation and export of meat. However, vacuum packaging has not proved to be a successful method for the retail packaging of meat, because of the purple deoxymyoglobin colour of the meat and the visible exudate that occurs in the packages^{3,30}. Meat that is packaged in a vacuum cannot be presented in the bright red oxymyoglobin state, which depends on the presence of a high concentration of O₂^{30,33}, or in the cherry red carboxymyoglobin state, which requires CO to be included in the MA.

One of the objections that has been raised against the use of CO as a packaging gas is the potential hazard it might represent to workers in meat plants. Although the use of pure CO for mixing in the plant would certainly pose such a risk, the delivery of 1% CO in a mixture with 99% N₂, which has been the practice of gas suppliers to the Norwegian meat industry, is recognized by the health authorities to be a very safe handling procedure.

MAs that contain 60–70% O₂ must be handled carefully, because they are explosive gas mixtures. Strict safety regulations apply to explosive gas mixtures, increasing the costs of equipment and packaging operations. The benefits of a CO mixture is that it carries no risk of explosion and therefore does not increase handling costs.

Despite the long-term knowledge of the many advantages of the use of CO as a component of MAs for meat, CO mixtures have not been adopted to any great extent by the global meat industry. In many countries, including the USA and countries within the EU, CO is presently not permitted for use in the MAP of meat^{3,4}. However, Norwegian food control authorities have not opposed the use of CO as a packaging gas at concentrations of up to 0.5%. As a member of the European Economic Agreement, Norway is expected to adapt gradually to EU food regulations, including those relating to gases for the packaging of foods. The Norwegian meat industry is therefore preparing an inquiry, to be directed at the Norwegian and EU food control authorities, for the continued use of CO in the MAP of red meats, which will be partly based on the toxicological

Table 5. Theoretical uptake of carbon monoxide (CO) in blood

Exposure method	CO intake in 1 h	CO intake in 8 h
Lungs (15 m ³ /g)	24 mg × 0.625 = 15.1 mg	9.2 mg × 5 = 46.0 mg
Meat (250 g, CO treated)	0.025 mg	0.025 mg

Consumers may evaluate the shelf life of packaged meat on the basis of its colour. A possible negative aspect of using CO in the MAP of retail meat is concern that consumers might misjudge the quality of a product, because its true microbiological status may be masked by its stable, cherry red carboxymyoglobin colour¹. However, consumers will be able to detect spoilage by the presence of off-odours. At the current low concentrations, <0.5%, CO *per se* seems to have no or only minor effects on bacteria and the shelf life of the meat. The combination of CO with a high concentration of CO₂, for example 60–70%, is necessary for microbiological control. Although MAP enables centralized packaging operations with quality control to be carried out, MAP alone cannot guarantee the shelf life of the product. Sufficient shelf life can be obtained only through the proper quality control of raw materials, production, packaging, chill chain and retail conditions.

Conclusions

Gas mixtures that contain a low concentration of CO, up to 0.5%, and a high concentration of CO₂, ~70%, have many advantages with respect to shelf life, colour stability, labour safety and costs. The use of CO at such concentrations does not present any toxic threat to consumers. Considering the benefits the Norwegian meat industry has experienced with CO gas mixtures over the past decade, potential exists for their wider application in the retail packaging of meat.

Acknowledgement

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The storage life of beef and pork packaged in an atmosphere with low carbon monoxide and high carbon dioxide

Oddvin Sørheim^{a,*}, Hilde Nissen^a, Truls Nesbakken^b

^aMATFORSK-Norwegian Food Research Institute, Osloveien 1, N-1430 Ås, Norway

^bNorwegian Meat Cooperative, Department of Research and Development, PO Box 360 Økern, N-0513 Oslo, Norway

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Abstract

Ground beef, beef loin steaks and pork chops were packaged in modified atmospheres of 0.4% CO/60% CO₂/40% N₂ and 70% O₂/30% CO₂. In addition ground beef was packaged in clipped chub packs, beef loin steaks were vacuum packaged, and pork chops were packaged in an atmosphere of 60% CO₂/40% N₂ with each pack containing an O₂ absorber. The packs were stored in the dark at 4 or 8°C for up to 21 days. Meat in 0.4% CO/60% CO₂/40% N₂ had a stable bright red colour that lasted beyond the time of spoilage. The storage lives in this gas mixture at 4°C, as limited by off-odours, were 11, 14 and 21 days for ground beef, beef loin steaks and pork chops, respectively. The 70% O₂/30% CO₂ atmosphere resulted in an initially bright red to red colour of the meat, but the colour was unstable and off-odours developed rapidly. The off-odours probably were caused by *Brochothrix thermosphacta*, which grew in all meat types, or by pseudomonads in ground beef. Meat stored in chub packs, vacuum packs or 60% CO₂/40% N₂ with an O₂ absorber developed off-odours and microflora similar to those of meat in 0.4% CO/60% CO₂/40% N₂, but with less acceptable appearances. These results show that a low CO/high CO₂ atmosphere is effective for preserving retail-ready meat. © 1999 Elsevier Science Ltd. All rights reserved.

1. Introduction

The main reasons for modified atmosphere packaging (MAP) of red meats for retail sale are to prolong the microbiological shelf life and to maintain an attractive red colour of the product. Modified atmospheres (MA) usually consist of carbon dioxide (CO₂) for inhibiting microbiological growth, oxygen (O₂) for enhancing colour and, occasionally, nitrogen (N₂) as a filler. The most common gas mixture for retail-ready meat contains approximately 70% O₂ and 30% CO₂, and gives the product an extended shelf life compared to air (Gill, 1996). The shelf life and colour stability of meat stored in this gas mixture is still limited. To obtain a stable red colour for the meat, low concentrations (<1%) of carbon monoxide (CO) can be introduced in the MA. Then, O₂ can be removed from the gas mixture and the concentration of bacteriostatic CO₂ can be increased. Anaerobic conditions extend the shelf life of meat considerably compared to air and O₂-enriched atmospheres (Gill & Molin, 1991). CO binds strongly to the meat

pigment myoglobin to form stable carboxymyoglobin which has a cherry red colour (El-Badawi, Cain, Samuels, & Angelmeier, 1964). Low concentrations of CO have little effect on the microflora of meat (Clark, Lentz, & Roth, 1976; Gee & Brown, 1978; Luño, Beltrán, & Roncalés, 1998).

The Norwegian meat industry has for the past decade been using a gas mixture of approximately 0.3–0.5% CO, 60–70% CO₂ and 30–40% N₂ in retail-ready packages of beef, pork and lamb. Packages with this gas mixture now have a 50–60% share of the domestic, retail, red meat market. The technological, hygienic and toxicological aspects of using CO in MA for meat have recently been reviewed with the conclusion that CO used in concentrations up to 1% does not present a toxic hazard to the consumer (Sørheim, Aune, & Nesbakken, 1997a). However, CO may mask spoilage, because the stable cherry red colour can last beyond the microbiological shelf life of the meat (Kropf, 1980).

The inclusion of CO in MA for meat is controversial. CO is presently not allowed in MA for meat in the USA and in the EU (Cornforth, 1994; European Parliament and Council Directive, 1995). However, Norwegian food control authorities have up to now not opposed

* Corresponding author. Tel.: +47-64-970100; Fax: +47-64-970333; E-mail: oddvin.sorheim@matforsk.no

the use of up to 0.5% CO in MA for meat. This would change with an adoption of EU food regulations in Norway. Consequently, the Norwegian meat industry is seeking amendments of current EU food regulations relating to the use of CO in MAP of red meats. If the use of CO should be disallowed, other means of maintaining the long shelf life and the attractive red colour of the meat will have to be sought.

The aim of the present experiments was to compare a commercial Norwegian CO/CO₂/N₂ mixture with alternative gas mixtures and packaging methods for their effects on the off-odour, microflora and colour of ground beef, beef loin steaks and pork chops stored at 4 or 8°C for up to 21 days.

2. Materials and methods

2.1. Preparation of meat

2.1.1. Ground beef

Twenty cow and bull carcasses of Norwegian Red Cattle, which weighed on average 275 kg, were electrically stimulated with 90 V and were chilled using programmed air temperatures between 12 and –5°C. Two days after slaughter the carcasses were deboned, and trimmings with 14% fat were ground through a 4 mm plate. The batch of ground beef was divided into 500 g portions.

2.1.2. Beef loin steaks

Loins (*m. longissimus lumborum et thoracis*) with ultimate pH values below 5.8 were deboned from 25 bull carcasses of Norwegian Red Cattle. These carcasses, which weighed on average 275 kg, were stimulated, chilled and deboned the same way as the carcasses used in the preparation of ground beef. The loins were vacuum packaged and aged for 11 days at 3°C. Thereafter, the loins were cut into steaks 2.5 cm thick, and were randomly assigned to retail packs which each contained two steaks.

2.1.3. Pork chops

Thirty pig carcasses of Norwegian Land Race, which weighed on average 75 kg, were blast-chilled. Four days after slaughter, bone-in loins were removed and crust-frozen in liquid N₂ at –50°C for 20 min to facilitate cutting of chops. The chops, which were 1.6 cm thick, were randomly assigned to retail packs which each contained two chops.

2.2. Packaging

Ground beef, beef loin steaks and pork chops were packaged in 0.4% CO/60% CO₂/40% N₂ (CO mixture) and 70% O₂/30% CO₂ (high O₂). In addition, ground beef was packaged in clipped chub packs, beef loin steaks were vacuum packaged and pork chops were packaged in 60% CO₂/40% N₂ with one Ageless® FX-

100 O₂ absorber (Mitsubishi Gas Chem. Co. Inc., Tokyo, Japan) in each pack (mixture with O₂ absorber).

The meat was packaged at a commercial meat plant within 2 h of grinding or cutting. Meat in the CO mixture, the high O₂ mixture and the mixture with O₂ absorber was packaged in an Ilapak Delta 2000 flow-packaging machine (Ilapak Machine Auto S.A., Grancia, Switzerland). The CO mixture was a blend of 1% CO/99% N₂ with 100% CO₂. The high O₂ mixture was used as a preblend. The mixture with O₂ absorber was a blend of 100% N₂ with 100% CO₂ (all gases, Hydrogas, Porsgrunn, Norway). The initial gas volume to meat weight ratio in the packs was approximately 1.5 to 1. The packs consisted of polyethylene trays (Færch Plast, Holstebro, Denmark) wrapped in Cryovac BDF 550 shrinking film (Cryovac, Milan, Italy) with an O₂ transmission rate of 19 cm³/m²/24 h/atm at 23°C and 0% RH. Chub packs of ground beef were packaged in a clipping machine (Poly-Clip, Frankfurt, Germany) using a red, fishingnet-patterned, polyethylene film (SFK, Vidovre, Denmark) with an O₂ transmission rate of 500 cm³/m²/24 h/atm at 23°C and 0% RH. Beef loin steaks were vacuum packaged in a Multivac 5100 thermo-forming machine (Multivac, Wolfertschwenden, Germany) using a terephthalate/polyethylene upper film and polyamide/polyethylene lower film with O₂ transmission rates of 10 and 16 cm³/m²/24 h/atm at 23°C and 0% RH, respectively (Danisco, Horsens, Denmark).

2.3. Storage and sampling of meat

Five samples were collected from the ground beef batch, beef loins and pork loins before packaging, for pH measurements and microbiological analyses.

The packaged meat was stored in dark chilling rooms at 4 ± 0.5 or 8 ± 0.5°C for up to 21 days at least until off-odours developed. Five packs were removed per product, packaging method, storage temperature and sampling day after the following storage times:

- ground beef: 2, 4, 6, 8 or 11 days;
- beef loin steaks: 3, 7, 10 or 14 days; and
- pork chops: 3, 7, 10, 14, 17 or 21 days.

2.4. Gas analyses

The atmospheres of packs with MA were analysed for O₂ and CO₂ immediately after packaging (approximately every tenth pack) and at sampling (all packs). O₂ was determined using a Toray LC 700-F gas analyser (Toray Engineering, Osaka, Japan) and CO₂ using a Toray PG-100 gas analyser (Toray). The threshold levels for the O₂ and CO₂ analyses were 0.05 and 1%, respectively. Gas samples of 10 cm³ were removed with a syringe through selfsealing patches on the packs.

2.5. pH

The pH measurements were made directly in the meat with an Ingold Xerolyt gel electrode (Mettler-Toledo A.G., Greifensee, Switzerland).

2.6. Odour

The meat was evaluated for odours by a three member trained panel between 0.5 and 1 min after opening of the packs. The off-odour scale used was: 1 = none, 3 = slight and 5 = extreme. Scores of 3 or below were considered acceptable.

2.7. Microbiology

Ten gram meat samples were collected from portions of the ground beef, and diluted in 90 g peptone water. A sample 25 cm² and 2–3 mm thick was removed from the surface of each beef loin or steak and pork loin or chop with a scalpel, and diluted in 100 ml peptone water. Each sample was macerated in a Stomacher for 1 min. Serial 10-fold dilutions of each Stomacher fluid were prepared, and 20 µl volumes of appropriate dilutions were plated in duplicate on the following media:

- plate count agar (PCA; Difco, Difco Laboratories, Detroit, MI, USA) for total viable counts;
- de Man, Sharpe and Rogosa agar (MRS; Oxoid, Unipath Ltd., Basingstoke, Hampshire, UK) adjusted to pH 5.7 for lactic acid bacteria (de Man, Rogosa, & Sharpe, 1960);
- streptomycin thallous acetate actidione agar base (STAA; CM 881 with selective supplement SR 151; Oxoid) for *Brochothrix thermosphacta*;
- pseudomonads agar base (CFC; CM 559 with selective supplement SR 103; Oxoid) for pseudomonads;

In addition, 1 ml portions of appropriate dilutions were plated in duplicate on petrifilm coliform count plates (3M Microbiology Products, St. Paul, MN, USA) for enumeration of coliforms and *Escherichia coli*.

Plates of PCA, MRS, STAA and CFC were incubated at 20°C for four days, and petrifilm plates at 30°C for up to 2 days, all aerobically. Counts were expressed as colony forming units (CFU) per g or cm².

2.8. Colour

A six-member trained panel evaluated the colour of the meat in intact packs under 1200 ± 200 lux Warmton Lumilux L36W/31 yellow-white light (Osram, Drammen, Norway). The colour was assessed on a scale where 1 = bright red (ground beef and beef loin steaks) or light bright red (pork chops), 2 = red (ground beef

and beef loin steaks) or light red (pork chops), 3 = slightly brown, grey or green, 4 = moderately brown, grey or green and 5 = extremely brown, grey or green (National Live Stock and Meat Board, 1991).

A Minolta Chroma Meter CR-300 (Minolta Camera Co., Osaka, Japan) with 8 mm viewing port and illuminant D₆₅ was used for measuring CIE *a** values (redness). The colour was measured directly at the meat surface within 1 min of opening of each pack.

Ground beef in chub packs was not included in the colour analyses because the red packaging film hides the colour of the product. With pork chops, the colour of only the *m. longissimus lumborum et thoracis* was analysed.

2.9. Statistics

Analysis of variance by Tukey's multiple comparisons test was performed using the Systat programme, version 6 (Systat Inc., Evanston, IL, USA).

3. Results

3.1. Gas composition

The initial O₂ concentrations in packs with the CO mixture and the mixture with O₂ absorber were all below 0.5% immediately after packaging. O₂ was not detected in these packs after 2 or 3 days storage. The level of O₂ in packs of high O₂ was reduced from the initial 70 to 60–65% during storage for up to 21 days. Concentrations of CO₂ in the packs were generally reduced by one fifth after 2 or 3 days storage, and were then stable (data not shown).

3.2. Storage life of ground beef

The time to develop off-odours was 2 to 3 days longer for ground beef stored in the CO mixture and in chub packs than in high O₂, and it was 4 or 5 days longer at 4 than at 8°C for all three packaging methods (Table 1). In high O₂, the total viable counts increased faster and were higher (*p* < 0.01) than for the other two types of packaging after 2 days at either 4 or 8°C [Fig. 1(a)]. The total viable counts were more than 90% lactic acid bacteria (data not shown). The high numbers of lactic acid bacteria in ground beef, up to approximately log₁₀ 8 CFU/g, caused a decrease in the pH value from the initial 5.7 to 5.2 after 6 days when the meat was stored in the CO mixture or chub packs at 8°C (data not shown). At 4°C, the pH value was reduced to 5.5 after 11 days in both those packaging systems. The numbers of *B. thermosphacta* increased, in meat in high O₂ [Fig. 1(b)]. In meat in high O₂ the numbers of pseudomonads increased up to approximately log₁₀ 7 CFU/g, but only to log₁₀ 5 and 6 CFU/g in

meat in the CO mixture or chub packs, respectively (data not shown).

Ground beef in the CO mixture had a stable bright red colour, as shown by both the low colour scores and the high a^* values [Fig. 1(c) and (d)]. Meat in high O_2 was significantly less red ($p < 0.05$) than meat in the CO mixture, with higher colour scores and lower a^* values at day 2 and at later storage times at both 4 and 8°C. The colour of meat in high O_2 deteriorated with time, significantly faster ($p < 0.01$) at 8 than at 4°C.

Table 1
Time for development of off-odours in different types of meat in various packagings at storage temperatures of 4 or 8°C

Product	Packaging ^a	Time of off-odour detection (days)	
		4°C	8°C
Ground beef	CO mixture	11	6
	High O_2	8	4
	Chub packs	11	6
Beef loin steaks	CO mixture	14	7
	High O_2	10	7
	Vacuum packs	14	7
Pork chops	CO mixture	21	14
	High O_2	14	7
	Mixture with O_2 absorber	17	10

^a CO mixture = modified atmosphere of 0.4% CO_2 /60% CO_2 /40% N_2 ; High O_2 = modified atmosphere of 70% O_2 /30% CO_2 ; Mixture with O_2 absorber = modified atmosphere of 60% CO_2 /40% N_2 with an O_2 absorber in the pack.

3.3. Storage life of beef loin steaks

At 4°C, off-odours developed 4 days later in beef loin steaks in the CO mixture and in vacuum packs than in high O_2 (Table 1). At 8°C, no differences in the development of off-odours were observed. Off-odours developed 4 to 7 days earlier in meat at 8 than at 4°C. The type of packaging did not significantly affect ($p < 0.05$) the total viable counts on the meat, but the counts were significantly higher ($p < 0.01$) at 8 than at 4°C after both 3 and 7 days of storage [Fig. 2(a)]. The numbers of *B. thermosphacta* were less than $\log_{10} 4$ CFU/cm² in meat in all types of packaging at all times, but were significantly higher ($p < 0.05$) on meat in high O_2 at 7 and 10 days than on meat in the CO mixture and in vacuum packs at equivalent times [Fig. 2(b)]. The numbers of pseudomonads did not exceed $\log_{10} 3.5$ CFU/cm² at any sampling time, and were not significantly affected ($p > 0.05$) by the type of packaging or the storage temperature.

The colour of the beef loin steaks in the CO mixture was stable bright red throughout storage at both 4 and 8°C, as shown by the low colour scores and high a^* values [Fig. 2(c) and (d)]. Steaks in high O_2 were also bright red with high a^* values at day 3, but these steaks discoloured gradually between days 3 and 10, significantly faster ($p < 0.05$) at 8 than at 4°C. Meat in vacuum packs was slightly discoloured with low a^* values throughout storage. The colour scores and a^* values of vacuum packaged steaks were not significantly affected ($p > 0.05$) by the storage temperature.

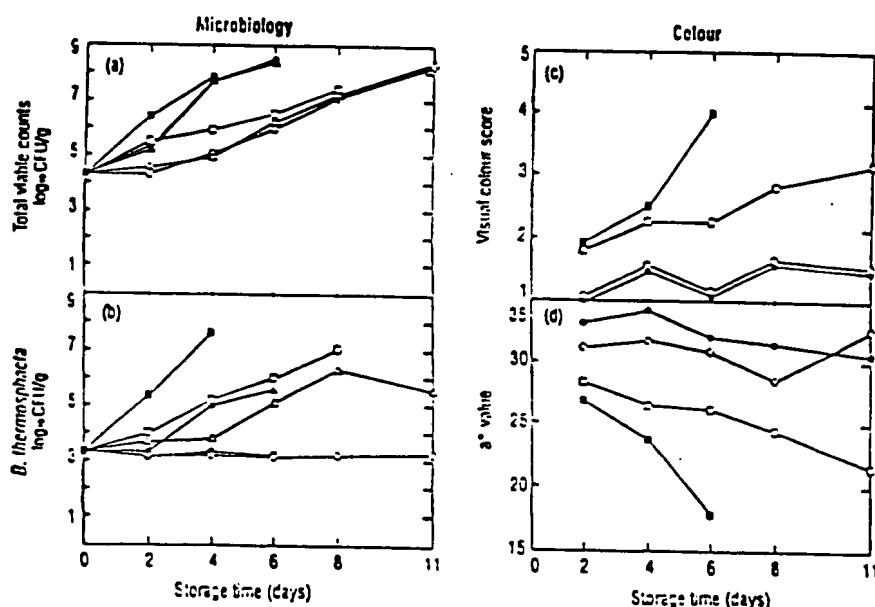


Fig. 1. Mean values ($n = 5$) for (a) total viable counts, (b) numbers of *Brochothrix thermosphacta*, (c) visual colour scores and (d) CIE a^* values for ground beef stored in 0.4% CO_2 /60% CO_2 /40% N_2 at 4°C (○) or 8°C (●), in 70% O_2 /30% CO_2 at 4°C (□) or 8°C (■), or in chub packs at 4°C (△) or 8°C (▲). Colour was assessed on a scale where 1 = bright red and 5 = extremely discoloured.

3.4. Storage life of pork chops

For pork chops, off-odours developed more slowly in meat in the CO mixture than in meat in the mixture with O₂ absorbers or in high O₂ (Table 1). Off-odours were detected 7 days earlier at 8 than at 4°C for chops in each type of packaging. The type of packaging did not affect the total viable counts on the pork chops [Fig. 3(a)]. However, the counts were greater on meat stored at 8 than at 4°C. The numbers of *B. thermosphacta* on chops in high

O₂ were significantly higher ($p < 0.01$) than on chops in the CO mixture or in the mixture with O₂ absorbers after 7 days at 8°C or 10 days at 4°C, and reached approximately $\log_{10} 6$ CFU/cm² [Fig. 3(b)]. The numbers of pseudomonads did not exceed $\log_{10} 3$ CFU/cm² on any of the pork chops.

The colour of pork chops in the CO mixture was light bright red with high a^* values throughout storage [Fig. 3(c) and (d)]. Chops in high O₂ were red at day 3, but discoloured during storage, significantly faster ($p < 0.05$) at

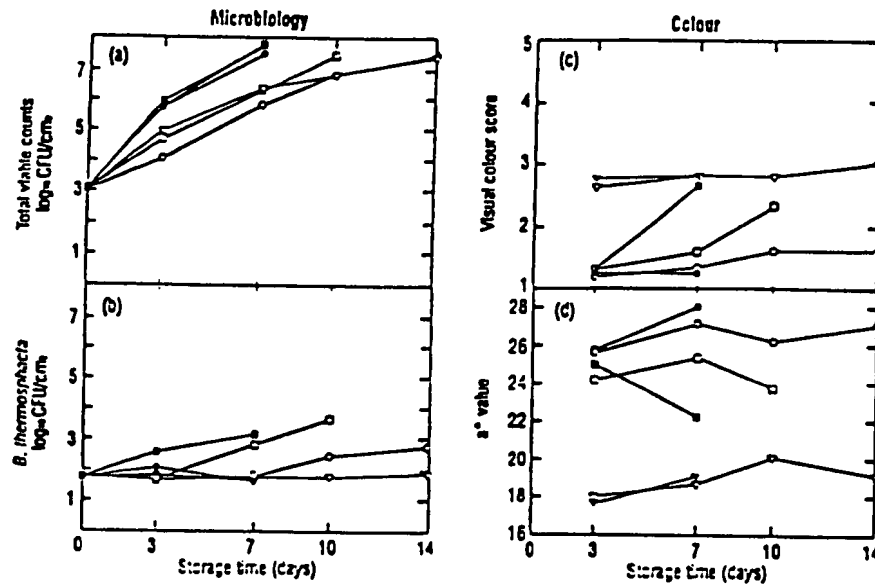


Fig. 2. Mean values ($n = 5$) for (a) total viable counts, (b) numbers of *Brochothrix thermosphacta*, (c) visual colour scores and (d) CIE a^* values for beef loin steaks stored in 0.4% CO/60% CO₂/40% N₂ at 4°C (○) or 8°C (●), in 70% O₂/30% CO₂ at 4°C (□) or 8°C (■), or in vacuum packs at 4°C (▽) or 8°C (▼). Colour was assessed on a scale where 1 = bright red and 5 = extremely discoloured.

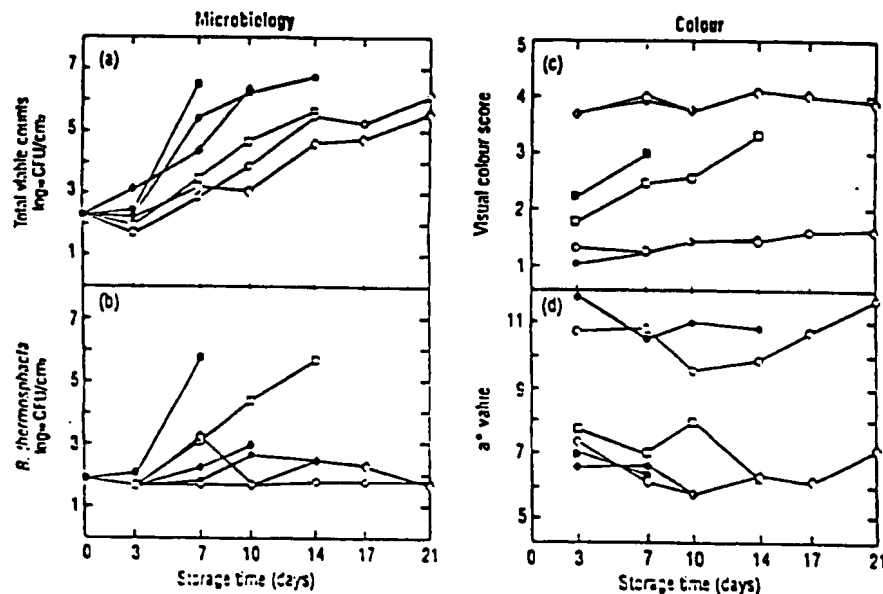


Fig. 3. Mean values ($n = 5$) for (a) total viable counts, (b) numbers of *Brochothrix thermosphacta*, (c) visual colour scores and (d) CIE a^* values for pork chops stored in 0.4% CO/60% CO₂/40% N₂ at 4°C (○) or 8°C (●), in 70% O₂/30% CO₂ at 4°C (□) or 8°C (■), or in 60% CO₂/40% N₂ with O₂ absorbers at 4°C (○) or 8°C (●). Colour was assessed on a scale where 1 = light bright red and 5 = extremely discoloured.

8 than at 4°C. Approximately 75% of the chops in high O₂ had black back bones at the time of sampling. Chops in the mixture with O₂ absorbers were moderately discoloured from day 3 to the end of storage. These chops had *a** values similar to those of chops in high O₂.

4. Discussion

4.1. Off-odour and microflora

The shelf life of the meat, as determined by the time to develop off-odours, was influenced by the packaging method, the storage temperature and the initial microbiological load on the meat. Storage of meat in the CO mixture, in vacuum packs or in chub packs gave the longest shelf lives. Meat stored in high O₂ generally developed off-odours 2-7 days earlier at 4 or 8°C than meat packaged in the other gas mixtures or by the other methods.

The differences in the rates of development of off-odours, as affected by the packaging method, were seldom related to any differences in numbers of total viable counts. However, the development of off-odours from the three meat types, especially ground beef and pork chops in high O₂, coincided with the attainment of high numbers of *B. thermosphacta*. For ground beef, storage in the CO mixture retarded growth of *B. thermosphacta* even more than storage in chub packs. At chill temperatures above 1°C, *B. thermosphacta* often causes spoilage of meat stored in high O₂ atmospheres (Dainty & Mackey, 1992). High concentrations of CO₂, removal of O₂ and low storage temperature inhibit the growth of *B. thermosphacta* (Gill, 1996; Nissen, Sørheim, & Dainty, 1996). Pseudomonads probably contributed to the off-odours of ground beef. Meat in high O₂ is often spoiled by *Pseudomonas* spp., but the growth of pseudomonads is retarded under anaerobic conditions (Dainty & Mackey, 1992; Gill, 1996). A shift in the metabolism of lactic acid bacteria under aerobic conditions can also produce off-odours (Nissen et al., 1996). In the present experiments, the numbers of coliforms or *E. coli* did not exceed log₁₀ 3 CFU/g or cm² in any samples. Therefore, these organisms probably did not contribute to off-odours.

For pork chops, the effect of CO on the microflora can be evaluated because the gas compositions of the CO mixture and of the mixture with O₂ absorber were identical, except for the inclusion of 0.4% CO in the former. Although a 4 day increase in the time to develop off-odours was observed with the CO mixture, there was no significant reduction in the microbiological counts. Luño et al. (1998) used 1% CO in high O₂ atmospheres and noted a delay in the onset of off-odours without any reduction in the numbers of psychrotrophic bacteria. However, Clark et al. (1976) found that the addition of

0.5-10% CO to N₂ atmospheres reduced the number of psychrotrophic bacteria and increased the odour shelf life of beef. For example, 1.0% CO in 99% N₂ increased the time to develop off-odours at 5°C from 18 to 24 days. The lack of such an effect of CO on bacteria in our experiments may be due to the use of 60% CO₂ overshadowing any effect of CO.

The use of CO makes it possible to dispense with O₂ and so to increase the CO₂ concentration in a MA to about 60%. Our data suggest that 0.4% CO probably has little or no direct effect on the growth of bacteria. Other studies have shown that increasing the CO₂ concentration from 20 to 100% increases the bacteriostatic effect of the gas, but the efficiency is highly dependent on low storage temperatures (Gill & Molin, 1991; Nissen et al., 1996). The high CO₂ concentration and absence of O₂ in the CO mixture will favour the growth of lactic acid bacteria, which usually cause a mild form of spoilage only late in the development of the spoilage flora (Gill, 1996).

The present experiments were performed at acceptable and abusive storage temperatures to assess the effects of temperatures commonly encountered in the distribution and sale of retail-ready meat. The storage temperature strongly affected the rates of growth of microflora and the time to develop off-odours. Consequently, independently of the packaging method, the shelf life of meat can be considerably extended by maintaining low temperatures in the chill chain (Gill & Molin, 1991; Nissen et al., 1996).

4.2. Colour

The CO mixture gave a stable bright or light bright red colour with consistent high *a** values for all three products, irrespective of the storage temperature. The initial level of residual O₂, up to 0.5%, did not adversely affect the visual scores and instrumental values for the colour of meat stored in the CO mixture.

CO binds to myoglobin and forms cherry red carboxymyoglobin (El-Badawi et al., 1964). This pigment is spectrally similar to the bright red oxymyoglobin which normally develops at the surface of fresh meat in air. Carboxymyoglobin is less readily oxidized to brown metmyoglobin than is oxymyoglobin, because of the strong binding of CO to the iron-porphyrin site on the myoglobin molecule (Lanier, Carpenter, Toledo, & Reagan, 1978; Wolfe, 1980). Consequently, CO in concentrations of 0.5-2.0% enhances and stabilizes a bright red colour of meat (Kropsf, 1980; Sørheim et al., 1997a). In a recent study, 1% CO in combination with 24 or 70% O₂ stabilized the colour of beef by reduced formation of metmyoglobin after storage at 1°C for up to 29 days (Luño et al., 1998). However, in a study of beef stored in a MA of 2% CO/78% CO₂/20% N₂, the colour of the meat was characterized as "too artificial" by

a sensory panel (Renner & Labadie, 1993). From our studies and experience from the Norwegian meat industry, 0.4% CO seems sufficient to produce a stable, attractive, bright red colour of meat.

All three meat types stored in high O₂ were bright red to red with high *a** values early in the storage periods, approaching the colour of meat in the CO mixture. As the microbiological counts of meat in high O₂ increased, the colour deteriorated, faster at 8 than at 4°C. Meat stored in a MA of high O₂ develops a thicker layer of oxymyoglobin than meat stored in air (Renner & Labadie, 1993). However, the oxymyoglobin gradually oxidizes to metmyoglobin, and the oxidation is faster at higher temperatures.

For cut bone, haemoglobin released from disrupted red blood cells in the marrow will accumulate at the surface and ultimately become black after the bone has been exposed to air or O₂ (Gill, 1996). Although bone blackening was not considered in the present visual colour evaluation, it can negatively affect the saleability of bone-in meat at retail display. The cut bones of pork chops stored in high O₂ blackened during storage, but this discoloration was not observed on bones in the CO mixture and the mixture with O₂ absorbers.

Beef loin steaks stored in vacuum packs were slightly discoloured with low *a** values at both 4 and 8°C. In these packs, meat juices were observed between the upper and lower films, but that did not influence the colour evaluations.

O₂ absorbers in packs with high CO₂ facilitate the removal of residual O₂ and maintain atmospheres free of O₂ during storage (Smith, Abe, & Hoshino, 1995). Low levels of residual O₂, above 0.01–0.15% for beef and 0.5–1.0% for pork, will inevitably discolour the meat (Penney & Bell, 1993; Gill, 1996; Sørheim et al., 1997b). When no CO is present in an O₂ depleted MA, it is essential to remove the residual O₂ as fast and completely as possible to avoid formation of metmyoglobin. In these experiments, pork chops stored in the gas mixture with O₂ absorbers were moderately discoloured during the whole storage period at 4 or 8°C. Despite the obvious visible differences, these chops had similar *c** values to the chops in high O₂. The discoloured surface made the chops unfit for sale, even in the early stage of storage. The present findings contrast with previous results, where the colour of porcine *m. longissimus thoracis et lumborum* was significantly improved by using O₂ absorbers in MAs of CO₂ with residual O₂ (Sørheim et al., 1997b). The present discoloration could be caused by incomplete use or function of the absorbers (Gill, 1996).

4.3. Benefits and disadvantages of a MA with low CO/high CO₂

An objection raised against using CO as a small component of a MA for retail-ready meat is the possi-

bility that the colour stability can exceed the microbiological shelf life, with the risk of masking spoilage of the meat (Kropf, 1980). Therefore, the consumer must evaluate the microbiological condition of meat in a CO mixture by off-odours. When a MA with CO is applied commercially, it is important to have a proper control of the hygienic condition of the meat raw materials and the chill chain temperatures.

CO used in concentrations below 1.0% does not present any hazard to the consumer, because consumption of meat packaged in such concentrations of CO will result in only negligible levels of carboxyhaemoglobin in the blood of consumers (Sørheim et al., 1997a). By delivering CO in a 1% mixture with 99% N₂, which is the practice of Norwegian gas suppliers, CO is considered safe for use in the working environment. Other MAs with high levels of O₂, up to 70%, must be regarded as explosive gas mixtures, which must be used with appropriate precautions for safety (Luño et al., 1998).

The suitability of gas mixtures and packaging methods for red meats for retail display depends on their ability to both reduce spoilage and stabilize colour. Gas mixtures with low concentrations of CO and high concentrations of CO₂ provide a combination of a long microbiological shelf life and a stable, bright red colour of meat. Meat packaged in a MA with high O₂ can achieve an initial bright red colour, but the microbiological shelf life and the colour stability are both considerably lower than those of meat in the CO mixture. Using CO eliminates the need to have O₂ as a component of the MA. Other MAs and packaging methods, like high CO₂ with O₂ absorbers, chub packs and vacuum packs may give a shelf life comparable to that of the CO mixture, but with a less acceptable colour or appearance of the meat. Thus, there appears at present to be no fully satisfactory alternative to the CO mixture used in packaging of retail-ready red meats in Norway.

Acknowledgements

The financial support of this study from the Research Council of Norway is highly appreciated. Vestfold-Buskerud Slakteri A/L, Sem and Hydrogas AS Utviklings-senter, Porsgrunn, are greatly thanked for packaging of the meat. We appreciate the gift of Ageless® O₂ absorbers from Cryovac Europe, Norderstedt, Germany. The technical staff and Per Lea (statistics) at MATFORSK are thanked for their skilful assistance in the study.

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SWEDISH MEATS

[logo]

Date
April 9, 1999

Ref. No.
33

Reference:
Lars Wedén, (+46) 8-725 81 03

Date
Ref. No.

Svein E. Skorstad
Norsk Kjøtt [Norwegian Meat]
P. O. Box 60 Refstad
N-0513 Oslo 5 NORWAY

RECEIVED:	<i>April 14, 1999</i>
CASE WORKER:	<i>K. Framstad</i>
FILE NO.:	<i>561</i>
J. NO./DOC. NO.:	<i>99/00721</i>

Brother:

Please refer to our conversation at the Scandinavian Butchers' Association meeting on March 19 regarding CO as a packaging gas. Here at Swedish Meats we are committed to creating opportunities to increase the meat packaging done by the manufacturer in Sweden. An approval of CO as a gas for use in foods would entail significantly greater possibilities of brand profiling of meat products in the future, which could help improve our service to the retailers. Moreover, efficient factory packaging could even reduce costs across the entire distribution chain ending with the consumer.

We therefore support Norsk Kjøtt's proposal to submit a joint application to the EU Commission for the addition of CO to EU's list of additives. The plan is to submit a joint application in June 1999.

Sincerely,

Swedish Meats

[signature]
Lars Wedén

DANSKE SLAGTERIER [Danish Bacon and Meat Council]	Axelborg, Axeltorv 3 1609 Copenhagen V Denmark Tel. (+45) 33 11 60 50	Fax (+45) 33 11 68 14 Telex 22 975 ds dk Telegram "danslagt"	[Logo]
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February 11, 2000
Tel. (+45) 33 73 25 68

Norsk Kjøtt [Norwegian Meat]
att: Jorunn Vormeland
PB 360 Økern
N-0513 Oslo
Norway

[Stamp:]Received: *February 14, 2000*
Case Worker: *JVO*
File No.: *562*
J. No./Doc. No. *00/00204*

Use of CO as Packaging Gas

In Norway, CO has so far been used in very minute quantities together with other packaging gases for gas packaged cuts of meat and ground meat. This packaging gas has not been approved in the EU yet, and has not been used in Denmark this far.

CO and the other packaging gases help ensure the storage life and color stability of fresh meat, which are important for central packaging and distribution of meat. The trend in Denmark is toward increased central packaging of fresh meat, since this is both efficient and ensures high microbiological quality. It would therefore be interesting to take advantage of the positive Norwegian experiences with the use of CO in this country, as well.

Published research shows that the risk of growth of a range of pathogenic bacteria is the same or reduced when using CO in combination with the traditional packaging gases. The use of this gas can thus help improve food safety (Food Microbiology and Food Safety into the Next Millennium, Proceedings of the Seventeenth International Conference of International Committee on Food Microbiology and Hygiene, Netherlands, 1999).

The use of CO in the given concentration of 0.3–0.5% should not represent any toxicological risk to consumers. CO is generally supplied in a concentration of 1% in a mixture with either N₂ or CO₂ and does therefore not represent any workplace hazard to operators during the packaging process.

We are not aware that the use of CO was discussed in the process of drafting Directive 95/2/EU of February 20, 1995 concerning additives other than colorings and sweeteners. This may be due to the fact that no country had expressed any interest in using this kind of gas at the time.

Since there are advantages to the use of CO as a packaging gas, as mentioned, and since there are no negative effects to either consumers or packaging operators, DANSKE SLAGTERIER can support an

application to the EU Commission to include CO on the list of approved additives, possibly limiting the amount.

Sincerely,

DANSKE SLAGTERIER

[Signature]

Anne Birgitte Lundholt

(Managing Director)

000145

(10)



Asociación de Industrias
de la Carne de España

TELEFAX

To: Dirk Dobbelaere
Secretary (CLITRAVI)

Subject: CO gas at meat packaging gas

Date: 14 of February 2000

Nº of pages: 1

Dear Dirk:

After reading the scientific documents that Mr. Truls Nesbakken handed out in the last T&L working group meeting. We will support the Norwegian proposal for authorization CO gas as a packaging gas within the UE.

We look forward to hearing from you soon.

Yours sincerely,

Miryam de Miguel
Dpto. Calidad-AICE

For info
To Norwegian CLITRAVI member
From CLITRAVI

21.2.2000

Truls Nesbakken
Fagsentret for kjøtt
PB 396 Økem
0513 Oslo
Norge

Use of CO as packaging gas for meat and meat products

After going through the scientific documents sent to us and having own projects supporting the results, our institute is ready to support the Norwegian proposal to allow CO gas as a packaging gas in EU.

Finnish Meat Research Institute



Markku Raevuori
Managing Director



Raymond Tuominen
Laboratory Director

The Use of CO as a Packaging Gas for Fresh Meat.

By Magne Yndestad

A previous report on the use of CO as a packaging gas concluded that there is unsatisfactory documentation on factors such as the development of pathogenic bacteria in the gas mixture in question (0.4% CO/60% CO₂/40% N₂). The Norwegian Research Center for Meat forwarded recent and complementary documentation on September 13, 1999.

Following scientific review, the content of this documentation can be summed up as follows:

Bacteriological Conditions

Numerous studies have been undertaken regarding trial storage involving concentrations of CO₂ in a range consistent with the "Norwegian" mixture (60-75% CO₂). Moreover, there are articles documenting the bactericidal effect of various other concentrations of gas mixtures containing CO₂. The conclusion to these trials is the following:

The low CO concentration (<0.5% CO) has no apparent effect on bacterial flora in products packaged with gas. This also holds true for N₂ (filler gas).

Concentrations of CO₂ below 5% may stimulate the growth of certain types of bacteria. Between 5 and 50%, we see an approximately linear inhibiting effect. This effect is somewhat significant, since the inhibition of growth of the sensitive flora is as much as 50% at 10% CO₂. The documented effect of CO₂ in high concentrations primarily applies to the psychrotrophic flora, including the most important spoilage bacteria.

As for the pathogenic bacteria, scientific literature in general points to the same tendency, i.e. inhibition of growth at both 4°C and higher temperatures (e.g. 10°C).

In comparison to other packaging methods or gas mixtures used, the mixture in question seems favorable both in terms of storage life and in terms of the relevant pathogens.

Following the last round of applications, The Norwegian Research Center for Meat has performed a relatively extensive study on freshly ground meat packaged in 0.3-0.5% CO/60-70% CO₂ and 30-40% N₂. Various pathogens, such as *E. coli* O157:H7, *Listeria monocytogenes*, and *Yersinia enterocolitica* were tested in this trial. The Research Center has evaluated factors such as the important possibility that the strong suppression of the general psychrotrophic flora may favor certain pathogens, which will not be inhibited to the same degree. The main conclusion, however, is that the aforementioned pathogens are inhibited both at 4°C and 10°C. Comparing the CO packaging method to packaging employing a high concentration of O₂ or vacuum, shows that the risk for growth of the applied pathogens is identical or lower when packaging with CO.

The Research Center has studied the circumstances concerning salmonella bacteria and the gas mixture in question in the same products. Since none of the cultures grew at +4°C, studies were only undertaken at 10°C.

In this case, storage with packaging gas containing CO performed worst with regard to *S. dublin*, *S. enteritidis* and *S. diarizonae*, as a relatively strong growth occurred following Day 2. *S. typhimurium* too had considerable growth, although "sausage" packaging scored lower.

This is completely in line with what is known about a whole range of salmonella bacteria in foods, i.e. that they hold up very well when competing with other bacteria, and also grow very well at temperatures around 8-10°C.

These facts emphasize the importance of cooling regardless of what packaging method is chosen.

Sensorial Circumstances

The last report pointed to the particular fact that the CO packaged meat could retain a fresh red color for days after spoilage set in. Hence, the consumer cannot see whether the meat he or she buys is spoiled, as opposed to fresh meat packaged in other types of gas packaging.

The Research Center notes that when opening a package, the consumer will detect any spoilage odor, and hence not eat the product. This may be true, but it is a fact that many people won't react to any incipient decay when the products looks completely "fresh." However, the packaging method for which approval is sought is meant for fresh meat that will be treated with heat prior to use. This is an additional safety factor that is important in a comprehensive evaluation.

Conclusion

The first bacteriological/sanitary statement made was based on the documentation available at the time. The new data and other relevant information from scientific literature indicate that there is sufficient evidence that the use of CO as a packaging gas as described in the application won't result in any increased risk of transmittal of food-borne diseases among consumers.

[Handwritten:]

From the report "Fresh Meat in Consumer Packaging" with modified gas containing CO₂ [illegible]

IV. Report by Tore Aune: "Fresh Meat in Consumer Packaging – A Toxicological Evaluation of the Use of up to 0.5% CO in a Gas Mixture."

FRESH MEAT IN CONSUMER PACKAGING – A TOXICOLOGICAL EVALUATION OF THE USE OF UP TO 0.5% CO IN A GAS MIXTURE

By Tore Aune

Carbon monoxide (CO) is a colorless gas that is primarily generated by incomplete combustion of organic material. The background concentration of CO in the atmosphere is approximately 0.01-0.09mg/m³ (0.009-0.08 ppm), while the concentration in larger cities may exceed 50mg/m³ as an 8 hour mean, depending on traffic.

General Health Effects

CO attaches to the iron of the hemoglobin in the red blood cells during generation of carboxyhemoglobin (COHb), and can thus affect the transport of oxygen in the blood and the supply of oxygen to the tissues. Compared to its affinity to oxygen, hemoglobin has approximately 240 times greater affinity to CO. CO also attaches to myoglobin, cytochromes, and some other enzymes, but these reactions are considered less important than the formation of carboxyhemoglobin (WHO 1979). The health impact on humans is mainly restricted to effects on the cardiovascular system, the nervous system, and certain types of proteins and cells in the bloodstream, as well as effects on embryos (SFT 1992).

The carboxyhemoglobin percentage (COHb %) is a function of the CO concentration in the inhaled air, the exposure time and the level of physical activity (Coburn et al., 1965) (see Table 1). A CO exposure resulting in a COHb concentration above 2% in the bloodstream of the most sensitive individuals (cardiovascular patients) has been shown to give symptoms of localized oxygen deficit and chest pains. Reduced work capacity occurs at a somewhat higher COHb%, and persons suffering from angina can tolerate less strain before an attack occurs. No health effects have been detected in healthy adults at COHb concentrations below 5%.

Table 1: Blood carboxyhemoglobin percentage as a function of CO concentration in air, exposure time and different degrees of physical activity (Coburn et al., 1965):

Exposure		COHb%		
CO Conc.	Time in Hours:	At rest	Moderate Activity	Strenuous Activity
10 mg/m ³	8	1.3	1.4	1.4
25 mg/m ³	1	1.0	1.5	2.0
40 mg/m ³	1	1.3	2.2	2.9

CO attachment to the hemoglobin is reversible. The half-life at ventilation at rest is approximately 4.5 hours.

A small amount of CO is continually formed in the body as a result of the decomposition of substances such as hemoproteins. This results in a COHb% of approximately 0.5. The uptake of CO through inhalation comes in addition to that. The average COHb level in non-smokers is estimated at 1.2-1.5%, while the level is 3-4% in smokers.

Survey of Health Effects Associated with CO Exposure

The negative health effects of CO are due to the fact that CO competes with oxygen for points of attachments on the hemoglobin molecule. Moreover, the release of oxygen in the tissues is reduced (WGHO 1987). Myoglobin is closely related to hemoglobin. It stores oxygen and promotes the diffusion of oxygen to muscle cells. In cardiac and skeletal muscles, myoglobin binds CO with an affinity that is 30-50 times higher than the corresponding affinity for oxygen. No reported studies have shown that the binding of CO to myoglobin can cause any health effect at a COHb level of 4-5%.

Uptake and liberation of CO occur at a relatively slow pace (hours), which means that short-time exposure to elevated CO levels will not result in any noticeable increase in the COHb level. SFT report No. 92/16 (1992) includes an overview of the correlation between blood COHb levels and health effects (Table 2).

Table 2: Correlation between blood carboxyhemoglobin levels and health effects (SFT 1992):

COHb%	Observed Effects in Humans:
50 and above	Unconsciousness, lethal when untreated.
30 and above	Headache, dizziness, nausea, and vomiting.
10 and above	May be lethal to cardiovascular patients. Headache in healthy individuals.
5 and above	Reduction of peak oxygen consumption under strenuous activity in healthy individuals.
5 and above	Impaired vision, learning ability and fine motor response.
5 and above	Exposure during pregnancy may affect the embryo.
2.9 and above	Individuals suffering from angina can tolerate less strain before an attack occurs.
2.3 and above	Reduced capacity for physical work, especially stamina.
2 and above	Possible reduced ability to concentrate and pay attention.
2 and above	Symptoms of localized oxygen deficit and incipient chest pains in cardiac patients.

The literature in the field does not seem to indicate that health effects have been proven in healthy adults exposed to CO resulting in a blood COHb concentration of less than 5%.

However, the data indicate that a COHb level of 2-3% may have negative effects on sick and sensitive individuals, such as people suffering from cardiovascular diseases.

Exposure to CO through the Air

With regard to CO as an air pollution factor, a team of Norwegian experts (SFT 1992) suggested air quality criteria at CO concentrations resulting in a maximum of 1.5% COHb during light physical activity (including the CO produced endogenically). The correlation between CO concentration, activity level, and exposure time in order not to exceed 1.5% blood COHb is shown in Table 3.

Table 3: Calculation of CO concentrations in the air resulting in a COHb level of 1.5%, including endogenic CO production (SFT 1992):

Exposure Time:	CO Concentration, mg/m ³		
	At Rest:	Moderate Physical Activity:	Strenuous Physical Activity:
15 min	170	80	52
30 min	86	42	29
1 hour	48	24	18
8 hours	11.5	9.2	9.2

Exposure to CO through Consumption of Fresh Meat Treated with a Gas Mixture

There is a paucity of information in scientific literature concerning exposure to CO through the consumption of fresh meat treated with a gas mixture containing CO. One of the most interesting references in this regard is a 1954 publication by A. L. Tappel et al., which is unfortunately not easily accessible. However, their work has been cited in other publications, e.g. in the study by Clark et al. (1976): Tappel et al. considered a US industrial sanitary norm for CO of 50 ppm (8 hours/day), and found that such exposure would result in a blood COHb level over a longer period of time that is approximately 14 times higher than the temporary increase caused by consumption of approximately 225 g meat, provided that the myoglobin and hemoglobin in the meat are saturated with CO, and that 100% of CO from this source is transferred to the blood of the consumer (an estimate representing a hypothetical worst-case scenario). According to the authors, such treatment of meat will thus cause only a very minor effect in comparison to what is considered the safety limit, even when assuming maximum uptake of CO. Watts et al. (1978) exposed beef to a gas containing 1% CO for 3 days, and found that this resulted in a CO saturation of approximately 30% of the myoglobin. CO was lost under such storage conditions, with a half-life of approximately 3 days. After cooking, the CO concentration in the meat decreased to

below 0.09 ppm (equivalent to approximately 0.1 mg/kg). Maximum loss after cooking (on burner at 195°C) amounted to approximately 85%.

Comparison of CO Exposure through Air and Meat (CO Treated)

There is little data available for such a comparison, but a rough overview nevertheless provides some points of reference. An adult inhales 10-20 m³ air per 24 hours (depending on the activity level). This is the equivalent of 0.42-0.84 m³ per hour (or 3.36-6.72 m³ per 8 hours).

To stay within a maximum blood COHb level of 1.5%, the CO concentration in the air must be 24 mg/m³ for 1 hour at moderate physical activity, at 9.2 mg/m³ for 8 hours (according to Table 3). In comparison, the CO exposure is 0.1 mg/kg after consumption of 250 g of heated CO-treated meat that has been treated for 72 hours in a gas containing 1% CO (Watts et al., 1978). Table 4 shows a calculation of CO intake from the air and a meal of CO-treated meat.

Table 4: Comparison of CO intake from air within a range without any health impact and theoretical intake of CO through consumption of a meal of CO-treated meat:

Path of Exposure:	CO Intake, 1 hour:	CO Intake, 8 hours
Lungs (15 m ³ /24 hours)	24mg x 0.625 = 15.1mg	9.2mg x 5 = 46.0mg
Meat	0.025mg	0.025 mg

For CO balance between air and blood is only achieved after a considerable period of time (hours). The absorption of gases from the intestinal canal to the blood is probably considerably less efficient than from the lungs, where the tissue allows for maximum gas exchange between the alveoli and the bloodstream. This implies that intake of CO through meat probably won't cause any demonstrable increase in the blood CO level (in the form of COHb). And at any rate, the exposure from meat is much lower (approximately one thousand times lower) than through the airways, as shown in the calculations above.

According to the Norwegian Institute of Air Research (SFT 1992), the CO concentration in larger Norwegian cities is on average between 1 and 2 mg/m³ during the winter. Maximum hourly values have been measured to approximately 60 mg/m³, and maximum values for 8 hours to about 40 mg/m³.

Evaluation of Other Gases Used in Foods in the EU

EU's Research Committee on Foods (SCF) has not considered CO. However, the expert team has considered other gases (EUR 1981), such as carbon dioxide (CO₂) and nitrogen oxide (NO). In this connection, the committee employed the following evaluation method, which should be applicable for CO, as well:

CO₂: This compound is a natural product of metabolism, and people are constantly exposed to carbon dioxide from the atmosphere, food and drink. Compared to this exposure, the residual content from its use as an extraction agent is insignificant. Establishing an ADI for this compound is unnecessary. The committee considers this compound acceptable as an extraction agent. It is unnecessary to determine concentration values for the residue.

N₂O: The pharmacological and pharmacokinetic properties of this gas are well known from the extensive use of N₂O as an anaesthetic. Even though no data on residual content are available, such amounts are probably so minor that they are not hazardous to the consumer. The committee finds that it is unnecessary to establish an ADI, and considers the use of N₂O as an extraction agent acceptable.

Toxicological Evaluation of the Use of CO as a Packaging Gas for Meat

People are continually exposed to carbon monoxide, both by means of endogenic production and by inhaled air. Toxicologically, it is the amount of CO bound to the blood hemoglobin (the carboxyhemoglobin percentage) that determines any health effects. The very first effects in sensitive individuals occur at COHb concentrations from approximately 2-3%. To prevent possible health effects even in the most sensitive individuals, a team of Norwegian experts has suggested limits for CO in the air that do not result in COHb concentrations above 1.5%, including the endogenic production at 0.5%. The above-mentioned estimates indicate that even if all CO in the prepared meat is transferred to the consumer's blood, the CO concentration – even a temporary concentration – will remain well below accepted limits in air. From a health perspective, the use of CO in concentrations below 0.5-1 % for fresh meat thus represents no toxicological risk.

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ATTACHMENT 4

Final Report

EVALUATION OF BEEF STEAKS AND GROUND BEEF IN THE PACTIV ACTIVE TECH PACKAGING SYSTEM: EFFECTS OF CARBON MONOXIDE IN THE PACKAGE ATMOSPHERE

for

**Pactiv Corporation
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Prepared by:

Kathy Hachmeister, MS

Melvin Hunt, PhD

Weber Hall

Dept of Animal Sciences & Industry

Kansas State University

Manhattan, KS 66506

Phone: 785-532-1232

Fax: 785-532-7059

George Milliken, Ph.D

Dickens Hall

Dept of Statistics

Kansas State University

Manhattan, KS 66506

Phone: 785-532-0514

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PROJECT SUMMARY

The effects of carbon monoxide (CO) in Active Tech modified atmosphere packages (MAP) were determined for:

- A. Initial product color,
- B. Stability of color during display, and
- C. Relationships of color deterioration and microbial populations.

Steaks from three beef cuts (strip loin, tenderloin, and inside round steaks) and ground beef were packaged in a MAP certified gas blend (0.4% CO, 30% carbon dioxide and 69.6% nitrogen) and stored at 35° or 43°F for up to 35 days. Cuts then were removed from MAP and displayed at 34°F until their color was approaching consumer unacceptability. Color and microbial parameters were measured and compared to baseline data of comparable product exposed to oxygen but not CO.

A fundamental goal of this research was to determine if CO extended the color life of beef cuts and ground beef beyond their microbial soundness, i.e., did color mask spoilage.

CONCLUSIONS

- > The Active Tech MAP system containing CO in the gas blend produced products that were equally as red as products packaged with traditional oxygen permeable overwrap.
- > Improvement in visual appearance especially in the tenderloin and inner portion of the inside round steaks were observed on day zero of display and throughout display.
- > Color of products exposed to CO was a typical, bright red when the outer MAP bag was removed and products were allowed to bloom for 60 to 90 minutes.
- > Color declines for products stored in MAP with CO compared well to baseline products exposed to oxygen. Hence, a typical discoloration pattern was seen in both baseline and MAP studies.
- > Color life for tenderloin and inside round steaks (and to a lesser extent ground beef) was slightly longer than their baseline counter parts, especially when stored 35°F vs. 43°F.
- > Although microbial growth curves changed in slope and exponential growth based on the environment in the packages, bacterial growth was neither encouraged nor suppressed by the addition of CO to the MAP gas blend.
- > Aerobic bacteria and facultative anaerobes followed typical patterns of growth contingent upon the environmental conditions.
- > Effects of storage temperature (35° vs. 43°F) and increased storage time (21 or 35 days) resulted in typical redness decline, increase in off-odors and microbiological changes.
- > CO neither masked spoilage nor resulted in color life extension beyond the point of microbial soundness.

INTRODUCTION

Marketing of case-ready meats has moved beyond the concept stage to reality. This method of delivering meat to retailers is expected to be the predominate system within five years. Some of the largest retailers are already paving the way for this makes-sense marketing system.

Modified atmosphere packaging (MAP) systems are a necessity for case-ready meats because current retail meat over wrapping does not fulfill requirements for shelf life and other needs. Processors can choose either high-oxygen or low-oxygen MAP for retail-ready meats. Both systems rely upon the meat having certain functional properties needed to optimize delivery of cuts with excellent display color life and sound microbial quality.

In low-oxygen MAP, such as the Active Tech System of Pactiv Corporation, it is essential that the meat achieve a stable red color that extends throughout storage and display. This usually is accomplished by modifying the package atmosphere so that the meat pigment returns to its purple-red state (deoxymyoglobin). Then, at display, packaged cuts are re-exposed to oxygen (air) to re-form a bright-red color (oxymyoglobin). Some muscles can easily accomplish this function whereas other muscles have a difficult time – due principally to short comings of their inherent muscle chemistry. Thus, novel ways to aid in obtaining desirable color during storage and display would be beneficial.

Gas atmosphere composition plays a critical role in the functionality and efficacy of MAP systems for meat. The atmosphere affects one or more of the following: product appearance, shelf life, microbial and palatability issues, gas dynamics, purge, and myoglobin functionality. Typical atmospheres for low-oxygen MAP utilize carbon dioxide (CO₂) and/or nitrogen (N₂) prior to the meat being re-exposed to oxygen. Addition of small amounts of carbon monoxide (CO) to a CO₂ and/or N₂ atmosphere could aid in producing a more functional pigment color in MAP, especially in meat cuts known to have lower color stability. CO is well known for its ability to bind to myoglobin and form a bright, crimson-red colored pigment known as carboxymyoglobin. However, carboxymyoglobin is believed to stabilize meat color beyond its microbial shelf life. Consequently, consumers may not be able to rely on color as an indicator of quality at time of purchase. Research is needed to address the use of low levels of CO in a MAP system.

HYPOTHESIS AND OBJECTIVES

This research was based on the hypothesis that a small quantity (<0.5%) of CO combined with the typical gases of MAP (CO₂ and N₂) would produce meat color complimentary to the quality needs of a case-ready meat delivery system without compromising consumer quality issues. More specific objectives evaluated the effects of CO in the Active Tech System for:

- The initial color of intact muscles and ground beef – this objective addressed color differences between meat in MAP containing CO vs. packaging in O₂.
- The color deterioration of these products during display -- these data defined the color display stability of meat in MAP containing CO vs. packaging in O₂.
- The microbial profile of the meat stored with or without mild temperature abuse – this portion provided information about microbial growth with CO in MAP relative to the time-honored relationship between color deterioration and spoilage.

EXPERIMENTAL PROCEDURES

This project involved two phases. The **Baseline Display Study** characterized the color and microbial traits of selected cuts and ground beef using typical oxygen-permeable packaging under typical retail display conditions. The **MAP Display Study** utilized the Pactiv Active Tech Packaging System in combination with a unique, certified gas blend (0.4% CO, 30% CO₂ and 69.6% N₂) in the package atmosphere during storage conditions (pre-display).

The outer MAP bag was removed and the products were displayed in the same manner as the baseline samples. All data from the MAP Portion were compared to the Baseline product.

RAW MATERIALS:

Twelve beef strip loins (NAMP #180 containing the *Longissimus* muscle), 18 tenderloins (NAMP #189A containing the *Psoas major* muscle), 12 inside rounds (NAMP #169A containing the *Semimembranosus* muscle), and 6 batches of ground chuck (80% lean) were obtained from a commercial source (PrairieLand Processors, Inc., Kansas City, KS) at four to six days postmortem. Vacuum packaged subprimals and trim that were received at the KSU Meats Laboratory had an internal temperature of 34°F and had never been frozen. Prior to product preparation, subprimals were stored at 34°F. This product was allocated to 6 replications (2 each of the strip loins and inside rounds and 3 tenderloins constituted a replication).

PRODUCT PREPARATION AND PACKAGING:

One inch thick steaks cut from each subprimal and ground beef formed into about one-pound blocks (Beef Steaker, Model 600, Hobart Corp., Troy, OH) were placed on Styrofoam trays (17S for strip loins, 4P for inside rounds, 1 for tenderloins, and 2P for ground beef) containing an absorbent pad (Ultra Zap Soakers, Paper Pak Products, La Verne, CA). Product was overwrapped with polyvinyl chloride (PVC) film (23,000ccO₂/m²/24hrs; Filmco MW4, LinPac, UK or Omnifilm 4P, Huntsman, Salt Lake City, UT) using a mechanical wrapper (Filmizer Model CSW-3, Hobart Corporation, Troy OH) and was assigned randomly to either a **Baseline Display Study** using only PVC-wrapped packages or a **MAP Display Study** using the Active Tech System of Pactiv Corporation. Trays for MAP were placed individually in barrier bags (4.5ccO₂/m²/24hrs: NXE 1-300, Alec Enterprises, Burnsville, MN) along with an oxygen absorber (MRM-200, Multisorb Technologies, Buffalo, NY) activated using Pactiv Active Tech Activator No.1. Barrier bags were evacuated, flushed with a certified gas blend containing 0.4% CO, 30% CO₂, and 69.6% N₂, and sealed (Freshvac Model A300, CVP Systems, Inc., Downers Grove, IL).

TREATMENTS:

Baseline Display Study: Twelve packages of ground beef and one steak ($\leq 1/8$ " fat trim) from each subprimal (12 strip loins, 12 inside rounds, 18 tenderloins, and the 6 batches of ground beef), were evaluated in a baseline study to establish the color and microbial parameters for meat never in MAP and exposed only to atmospheric oxygen. These packages were placed in display about 4 hours post-packaging (see display and measurement details below).

MAP Display Study: To test the effects of CO in MAP, one package of each product from each of 6 replications was selected at random for assignment to all possible combinations of two storage temperatures (35 and 43°F) and three storage times (7, 14, and 21 days for ground beef and 7, 21, and 35 days for steaks). The lower temperature represented reasonably good industry practice, and the higher temperature represented a mildly abusive storage conditions. The storage times represented current industry practice.

Prior to display (post-MAP), the O₂ and CO₂ levels in the outer barrier bags were measured using a MOCON head space analyzer (Pac Check™ Model 650, MOCON/Modem Controls, Inc., Minneapolis, MN).

DISPLAY CONDITIONS:

Meat samples were placed in simulated retail display at $34 \pm 3^\circ\text{F}$ under 1614 lux (150 \pm 5 foot candles; Model 201, General Electric, Cleveland, OH) light intensity (Philips, 34 Watt, Ultralume 30) in open-top display cases (Unit Model DMF8, Tyler Refrigeration Corporation, Niles, MI). Cases were programed to defrost two-times per day at 12 hour intervals. Display case temperatures were monitored during display using temperature loggers (Omega Engineering, Inc., Stamford, CT). Display times varied based on product type, initial microbial loads, and storage conditions. Product was removed from display when the color score was deemed unacceptable by a visual panel (a color score of 3.5). Baseline products were displayed 7, 5, 4, and 3 days for strip steaks, inside rounds, ground beef, and tenderloins, respectively.

VISUAL COLOR EVALUATION:

Ten trained visual panelists evaluated color using a five-point scale where 1 = very bright red, 2 = Bright red, 3 = Slightly dark red or tan, 4 = Moderately dark red or tan, and 5 = Extremely dark red or brown. The cut-off score for consumer acceptable color was ≥ 3.5 .

Two portions of the inside round muscle were scored separately. The outer 1/3 portion (OSM) and the deep, inner 1/3 portion (ISM). The middle 1/3 area was not scored. The 10 panel scores were averaged for statistical analysis.

INSTRUMENTAL COLOR AND SPECTRAL DATA:

Samples were instrumentally analyzed for lightness (L*), redness (a*), and yellowness (b*) for Illuminant D-65 (daylight) using a HunterLab MiniScan Spectrophotometer (1.25 inch diameter aperture, Hunter Associates Laboratory, Inc., Reston, VA). Multiple readings (2 to 4 depending on cut size) were taken and averaged for statistical analysis on each cut at each testing period.

ODORS:

At the end of display, each package from the MAP Display Study was evaluated for off odors by two experienced panelists using a 5-point scale where 1 = no, 2 = slight, 3 = small, 4 = moderate, and 5 = extreme off odor. A score of 3.5 was assumed to be unacceptable to consumers.

MICROBIOLOGICAL PROCEDURES:

Microbial populations were estimated at the end of MAP storage (day 0 of display) and at the end of display (day of unacceptable color). For each post-display sample, a portion of the surface area (top surface) that had been exposed to light was excised. After each package was opened aseptically, two cores (ca 2 in²) were removed (approximately 1/8 inch depth), placed in a sterile stomacher bag, and blended two minutes with 0.1% peptone diluent. Serial dilutions of the homogenate were prepared in 0.1% peptone and appropriate dilutions were plated in duplicate on Aerobic Plate Count Petrifilm™ to determine total aerobic bacterial populations and on *E. coli* Count Petrifilm™ to estimate generic *E. coli* and total coliform bacterial counts. In addition, appropriate dilutions also were plated in duplicate on MRS agar to determine lactic acid bacterial populations. Aerobic Plate Count Petrifilm™ and *E. coli* Count Petrifilm™ (3M Microbiology Products, St. Paul, MN) were incubated at 90°F for 48 hours prior to enumeration. LAB populations were counted after 48 hours of 92°F incubation in a CO₂ chamber. Microbial detection limits for intact muscle and ground beef were 1.76 count/cm² and 5.0 count/gram, respectively.

pH:

pH was determined on intact muscle and ground beef samples collected on the day of production. Ten grams of sample were added to 100 mL of distilled water and blended for two minutes. A standardized pH meter with an electrode was used to measure pH according to the procedure outlined in the Handbook for Meat Chemists.

FAT AND MOISTURE:

Ground beef samples collected on the day of production were analyzed in duplicate for moisture and fat using AOAC procedures 985.14 and 985.15, respectively.

EXPERIMENTAL DESIGN AND STATISTICS:

The design was a randomized complete block with six replications. A replication consisted of 1 to 3 subprimals (number depended on the size of each cut). Steaks cut from the subprimals and separate batches of ground beef trim were randomly assigned to replication and the treatment combinations. Data were analyzed using analysis of variance and significant differences determined using least significant difference tests at $P < 0.05$.

SAMPLING TIMES/PARAMETERS MEASURED:

1. MAP Gas Composition for oxygen and carbon dioxide levels

- Subsample of several ActiveTech packages on production day (2-3 hours post-packaging) to verify gas composition being obtained
- End of MAP storage at two temperatures

2. Microbiology:

- Initial counts for subprimals and ground beef on the day of production
- End of MAP storage at two temperatures
- End of display

3. Visual Color:

- Initial color prior to display lighting
- End of MAP storage at each of two temperatures and after 60 to 90 min bloom at 34°F (equal to 0 time of display)
- Daily during display

4. Instrumental Color:

- Initial color = After packaging in PVC on production day for baseline data, minimal exposure to light
- End of MAP storage at each of two temperatures and after 60 to 90 min bloom at 34°F (equal to 0 time of display)
- Daily during display

5. Odor:

- At end of display (prior to microbial testing)

RESULTS AND DISCUSSION

The Baseline Study: A random selection of all steaks and ground beef packaged in PVC film were placed in display to serve as a baseline for color and microbiological comparisons. Products were expected to have the lowest microbiological load and ideal color stability using traditional packaging and display conditions for products exposed only to atmospheric oxygen. The inherent muscle chemistry responsible for good color life also was optimal. If the product exposed to CO were to have extended meat color life, then it will be compared to the baseline "control" with the "best" possible color.

Color Reference Points: The discussion below involves both visual and instrumental measures of color. Visual scores were considered the "standard" with instrumental color being discussed relative to its agreement or disagreement with the visual panel, ie, did the objective measurements confirm what the color panel saw. Visual scores of ≥ 3.5 were considered borderline acceptable. When samples reached this discoloration, they were removed from display. Normally, a^* values (higher values indicate more redness) are highly correlated to visual appraisal.

Inside round steaks typically are two-toned in color. The inner portion (ISM) is much less color stable compared to the outer portion (OSM). These portions were scored separately since one portion may have acceptable color while the other has unacceptable color that would be discriminated against by consumers resulting in the whole cut being judged

unacceptable in color. The effects of CO on this bi-colored muscle were needed to confirm that color was not excessively extended in either portion.

FAT AND MOISTURE, pH, AND INITIAL MICROBIAL LOAD:

Average fat and moisture contents of the ground beef were 19.5 and 61.6%, respectively. pH of both intact muscles and the ground beef ranged from 5.3 to 5.7. The initial aerobic plate counts and lactic bacteria counts for all products were relatively low and indicative of microbial quality of the raw materials and good sanitation. Furthermore, coliforms and *E. coli* were below the detection limit throughout the study.

GAS COMPOSITION AT END OF MAP:

At the end of MAP storage, each package atmosphere was analyzed for O₂ and CO₂ (Table 1). Only 6 (each from a different treatment combination) of 288 packages were removed from the experiment due to leakage.

INITIAL PRODUCT COLOR AND APPEARANCE:

The color of ground beef and steaks entering display (after MAP storage at 2 temperatures) was an attractive, typical red color. Although there were several significant differences in visual scores and a* values (Table 2 and Figures 1-10 at day 0) for product in CO vs. baseline cuts, the variation in color was usually within ± 0.5 of a color score. In general, the initial color of product exposed to CO was very similar to the color of steaks from the baseline display (never exposed to CO). When differences occurred, they were more related to either storage temperature or postmortem age of the product.

Panelists did not consider the color of product exposed to CO atypical. Cuts exposed to CO generally appeared more uniformly bright-red and would be expected to have high consumer appeal. These results were expected, as CO is known to preferentially form a ligand with the colored pigment (myoglobin) in meat resulting in an intensely red pigment known as carboxymyoglobin. At higher levels of CO (0.4% vs. 0.6 to 1%) than used in this experiment, meat color has been described as being an unusual crimson, bright-red color compared to the normal red of oxymyoglobin.

A critical next question was whether the carboxymyoglobin formed on the surface was more stable than the oxymyoglobin formed in baseline product. Further, did the carboxy

pigment deteriorate in a predictable way that consumers could continue to use visual color to judge freshness or potential spoilage.

COLOR DETERIORATION PROFILE:

Visual panel scores (Figures 1-5) and instrumental color (a^* values, Figures 6-10) clearly showed that product exposed to CO during MAP storage had color deterioration during display. As expected, visual scores increased (color deteriorated) and a^* values decreased (loss of redness) as days in display increased.

In several instances, color appeared to improve late in display – as indicated by a decrease in visual scores (see ground beef, strips loins and tenderloins at 43°F). These decreases were not a return of redness. Rather the apparent decrease resulted from removal of discolored packages the preceding period, leaving product with less overall discoloration remaining in the case.

In general, the color deterioration profiles followed an expected pattern. Namely, the freshest product (baseline packages) had the most stable, red color and the most days in display needed to reach borderline discoloration (Table 3 scores to 3.5) of all treatments. Exceptions occurred for the inside portion of the inside round and tenderloin products, where the product exposed to CO had slightly more stable color than the baseline product (Table 3). These two muscle areas are well known by retailers as having short color life. Thus, CO appeared to improve color life when the inherent muscle chemistry desired for color was limited.

For product from MAP, the longer the storage time, the faster the deterioration, especially at the higher storage temperature (Tables 2 and 3). For packages stored at 43°F, which was a mildly abusive temperature, color deterioration would be expected to accelerate. This phenomenon also is illustrated in Figures 1-10.

Changes in a^* values (and other instrumental measures of color not shown) followed the same pattern of color deterioration observed by the visual panelists. There was no evidence that color shelf life was unexpectedly lengthened by exposure of meat to CO in MAP. The question remaining is whether the color life of product in CO masked spoilage, ie, were microbial counts higher than expected based on the degree of discoloration?

COLOR DETERIORATION AND MICROBIAL GROWTH:

Baseline Display Study: Initial, pre-display microbiological data suggested that the raw materials were fresh and processed using good hygienic practices. For intact cuts, lactic acid bacteria, generic *E. coli*, and total coliform counts were below the detection limit of 1.76 CFU/in². Initial, pre-display APC for intact muscles ranged from 1 to 1.63 log₁₀ CFU/in². Post-display counts were higher ($P < 0.05$) than pre-display APC which was an increase in bacterial proliferation and typical deterioration. However, all product had sufficient microbes to be susceptible to spoilage.

Baseline products were pulled from display when the visual panel scores reached ≥ 3.5 . However, the APC did not exceed 5 log₁₀ CFU/unit as shown in Figures 11-14 and lactic bacterial did not exceed 6 log₁₀ CFU/unit as shown in Figures 15-18. Furthermore, off-odor scores for product at end of display (Table 3) ranged from no to slight off odor. Thus, color life in this base population did not exceed microbial soundness.

MAP Display Study: Similar trends in microbial growth occurred in post-displayed samples stored in MAP compared to baseline products. Microbial patterns for product deterioration are shown in Table 4 and Figures 11-18. Products stored under MAP at a slightly abusive temperature showed, as expected, a more rapid increase ($P < 0.05$) in microbial counts compared to samples stored at 35°F. For post-MAP (pre-display) and post-display samples, APC were higher at 43°F than 35°F (Table 4), and during the later days of storage at the higher temperature, differences were more obvious. Significant changes ($P < 0.05$) occurred in all cuts and ground beef with the exception of SM. Counts for the SM muscle were lower than expected and no significant changes occurring until day 35 of MAP storage. This suggests that quality products that have been handled in a sanitary fashion can be stored in the MAP system up to 35 days without comprising microbial quality. The APCs for intact strip loin and tenderloin steaks stored at 35°F were lower ($P < 0.05$) on all days of display on days 21 and 35 post-MAP than steaks stored at 43°F (Figures 12 and 14). Although products did not show a difference in APCs 7 days post-MAP, those products stored at the higher temperature (43°F) were more inferior 21 and 35 days post-MAP.

Did Color Mask Spoilage? Central to this research was to evaluate the idea that the color of CO treated meat might mask spoilage. Food scientists generally agree that meat

color is seriously discolored when microbial counts approach $\log 10^6$, and that off odors frequently appear at counts of 10^7 to 10^8 . Numerous studies of ground beef, frequently the product with the highest counts, show that consumer-purchased retail product often has counts of 10^5 to 10^8 .

Visual color scoring was considered as the "standard" for determining the time to remove products from display. Because the visual panel scores were the deciding factor for length of shelf life, the interdependence between visual color and APC, LAB, and odor were considered quite important.

Figures 19-21 show aerobic and lactic bacterial growth and odor scores at the end of display plotted against their corresponding visual color scores. All data observations were summed over storage temperature, storage time, and product type and plotted in one graph. If color masked spoilage, then there should be multiple points in the upper left quadrant of the plot, the area represented by unacceptable microbial counts and off odors but with acceptable color (i.e., scores <3.5). This did not occur with any frequency in any of the three plots. Thus, it does not appear that exposure of meat to CO during extended (up to 35 days at either 35° or 43°F) caused meat color to hide spoilage.

Table 1 - Carbon Dioxide (CO₂) and Oxygen (O₂) Levels in MAP Packages of ground beef (GB) and steaks from strip loins (LD), inside round (SM), and tenderloin (TL).

Meat Cut	Storage Temperature, °F	Storage Time, days	CO ₂ , %	O ₂ , %
GB	35	7	28.4	0
GB	43	7	28.7	0
GB	35	14	27.7	0
GB	43	14	28.3	0
GB	35	21	27.4	0
GB	43	21	28.0	0
LD	35	7	33.3	0
LD	43	7	34.2	0
LD	35	21	32.4	0
LD	43	21	31.8	0
LD	35	35	31.1	0
LD	43	35	28.5	0
SM	35	7	28.9	0
SM	43	7	29.7	0
SM	35	21	27.9	0
SM	43	21	27.3	0
SM	35	35	26.8	0
SM	43	35	24.6	0
TL	35	7	34.3	0
TL	43	7	34.8	0
TL	35	21	33.6	0
TL	43	21	32.3	0
TL	35	35	32.5	0
TL	43	35	29.2	0

Table 2 - Means for initial visual color and a* values for beef cuts exposed to carbon monoxide during storage at 35° and 43°F in Active Tech MAP vs. baseline cuts exposed only to oxygen.

Trait	Product	Baseline cuts	Time ^d in Active Tech MAP, days at 35° F		
			7	14 / 21	21 / 35
Initial Visual Color	GB	1.3a	1.6b	1.7b	1.8b
	LD	2.2b	2.5b	1.8a	2.2b
	ISM	1.8ab	2.0b	1.7a	2.0b
	OSM	2.6b	2.6b	1.9a	2.5b
	TL	1.9a	2.0a	1.9a	2.1a
Initial a* Values (redness)	GB	23.4a	25.6b	25.9b	25.6b
	LD	25.8a	25.7a	27.1ab	28.1b
	ISM	28.5a	26.9a	30.0a	29.4a
	OSM	27.4a	27.7a	29.8a	29.5a
	TL	23.6a	27.5b	30.0c	29.3c
			Time ^d in Active Tech MAP, days at 43° F		
Initial Visual Color	GB	1.3a	1.7b	1.8b	2.5c
	LD	2.2a	2.3a	2.1a	2.0a
	ISM	1.8a	1.8a	1.7a	2.4b
	OSM	2.6b	2.2a	2.2a	2.0a
	TL	1.9a	2.0ab	1.8a	2.2b
Initial a* Values (redness)	GB	23.4a	25.7b	25.1b	25.5b
	LD	25.8a	25.5a	28.7b	27.5b
	ISM	28.5a	28.7a	28.6a	27.5a
	OSM	27.4a	27.7a	30.2b	29.4ab
	TL	23.6a	27.8b	28.7b	26.4b

a-c Means in the same row with a different letter differ (P<0.05).

d Ground beef stored 7, 14, and 21 days, other muscles 7, 21, and 35 days.

Table 3 - Means for days to visual unacceptable visual color (score of 3.5) and odor at end of display for beef cuts exposed to carbon monoxide during storage at 35° and 43°F in Active Tech MAP vs. baseline cuts exposed only to oxygen.

Trait	Product	Baseline cuts	Time ^e in Active Tech MAP, days at 35° F		
			7	14 / 21	21 / 35
Days in display to unacceptable color	GB	3.6c	3.0b	3.0b	2.3a
	LD	6.2c	5.0b	5.2b	3.8a
	ISM	3.2a	4.8c	4.0bc	3.5ab
	OSM	4.8c	3.5b	3.4b	2.6a
	TL	2.6a	3.0b	3.2b	2.8ab
			Time ^e in Active Tech MAP, days at 43° F		
Days in display to unacceptable color	GB	3.6d	3.0cd	2.3b	1.5a
	LD	6.2d	5.0c	3.3b	2.3a
	ISM	3.2b	4.0bc	3.1b	2.0a
	OSM	4.5d	3.0c	2.4b	1.6a
	TL	2.6ab	3.0b	2.3ab	1.7a
			Time ^e in Active Tech MAP, days at 35° F		
Off-odor score at end of display	GB	1.5a	1.9a	2.8b	2.4ab
	LD	1.3a	1.3a	2.3b	2.3b
	SM	1.5a	2.2a	3.0b	3.0b
	TL	1.6a	1.2a	3.1b	3.3b
			Time ^e in Active Tech MAP, days at 43° F		
Off-odor score at end of display	GB	1.5a	3.3a	3.6a	3.9a
	LD	1.3a	2.9a	3.3ab	3.6b
	SM	1.5a	2.2a	3.4b	4.0b
	TL	1.6a	2.7a	3.3b	3.8c

a-d Means in the same row with a different letter differ (P<0.05).

^e Ground beef stored 7, 14, and 21 days, other muscles 7, 21, and 35 days.

^f Off-odor scale: 1 = none, 2 = slight, 3 = Small, 4 = Moderate, 5 = Extreme.

Table 4 - Means for aerobic plate counts (APC) on beef cuts exposed to carbon monoxide during storage at 35° and 43°F in Active Tech MAP vs. baseline cuts exposed only to oxygen.

Trait	Product	Baseline cuts	Time ^e in Active Tech MAP, days at 35° F		
			7	14 / 21	21 / 35
End of MAP storage APCs, log 10 cfu	GB	2.7a	2.6a	4.7b	5.5b
	LD	.7ab	0.2a	1.4bc	1.7c
	SM	1.0b	0.3a	0.3a	0.3a
	TL	1.3b	0.2a	2.6bc	3.1c
End of display APCs, log 10 cfu	GB	4.3a	4.4ab	5.6b	5.5b
	LD	1.4ab	0.4a	2.9bc	3.4c
	SM	0.6a	0.1a	0.6a	2.0b
	TL	0.3a	1.3b	3.5c	3.4c
			Time ^e in Active Tech MAP, days at 43° F		
End of MAP storage APCs, log 10 cfu	GB	2.7a	4.6b	5.8c	6.0c
	LD	0.7a	1.3ab	3.2c	5.1d
	SM	1.0b	0.1a	0.1a	2.8c
	TL	1.3a	1.6a	3.7b	4.0b
End of display APCs, log 10 cfu	GB	4.3a	5.8b	5.9b	6.1b
	LD	1.4a	1.3a	2.8b	5.3c
	SM	0.6a	0.3a	0.7a	2.5b
	TL	0.3a	3.3b	4.2b	4.6b

a-d Means in the same row with a different letter differ ($P < 0.05$).

e Ground beef stored 7, 14, and 21 days, other muscles 7, 21, and 35 days.

Figure 6
Ground Beef a^* Values (Redness) Deterioration
During Display Following Storage

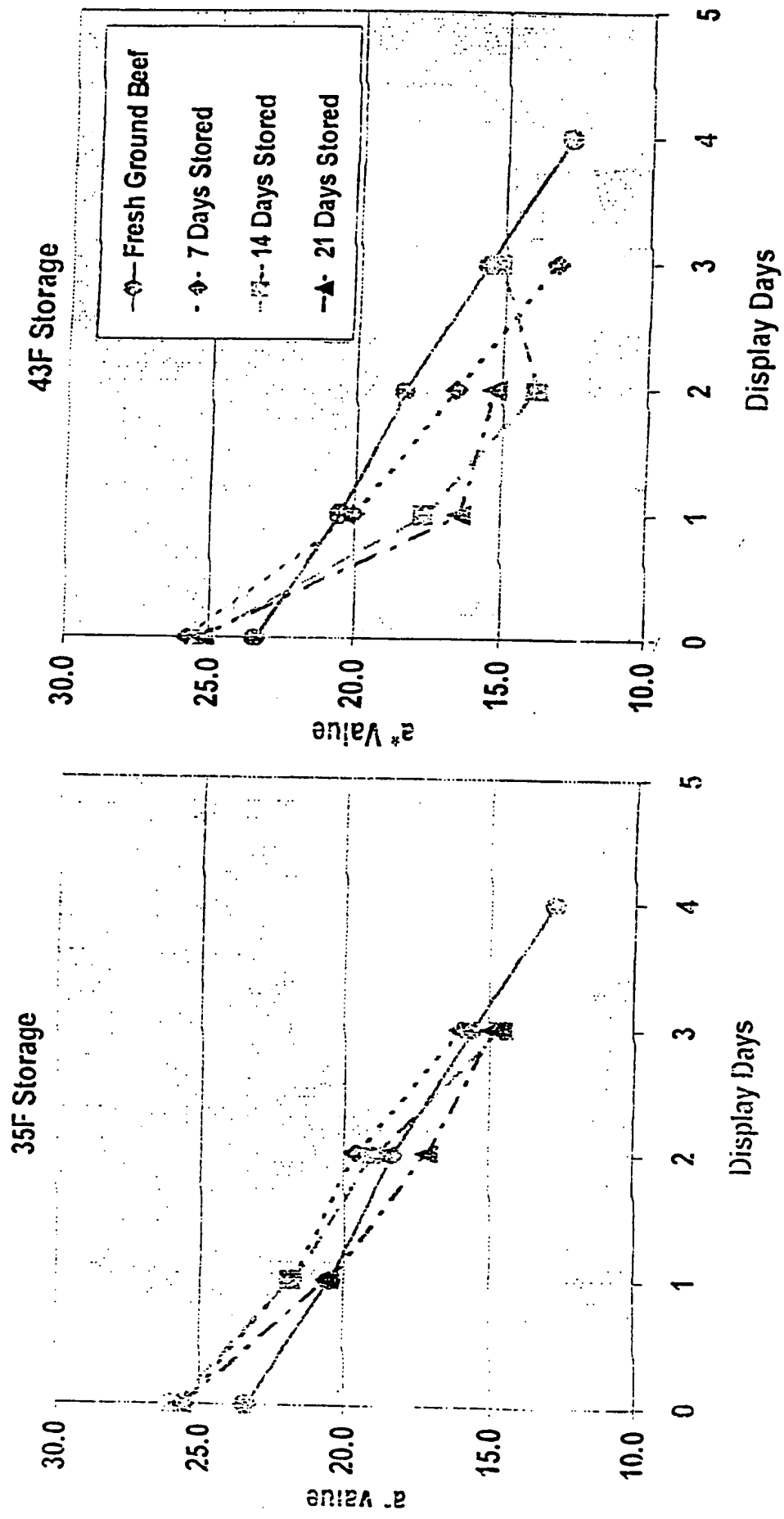


Figure 7
Strip Loin a^* Values (Redness) Deterioration
During Display Following Storage

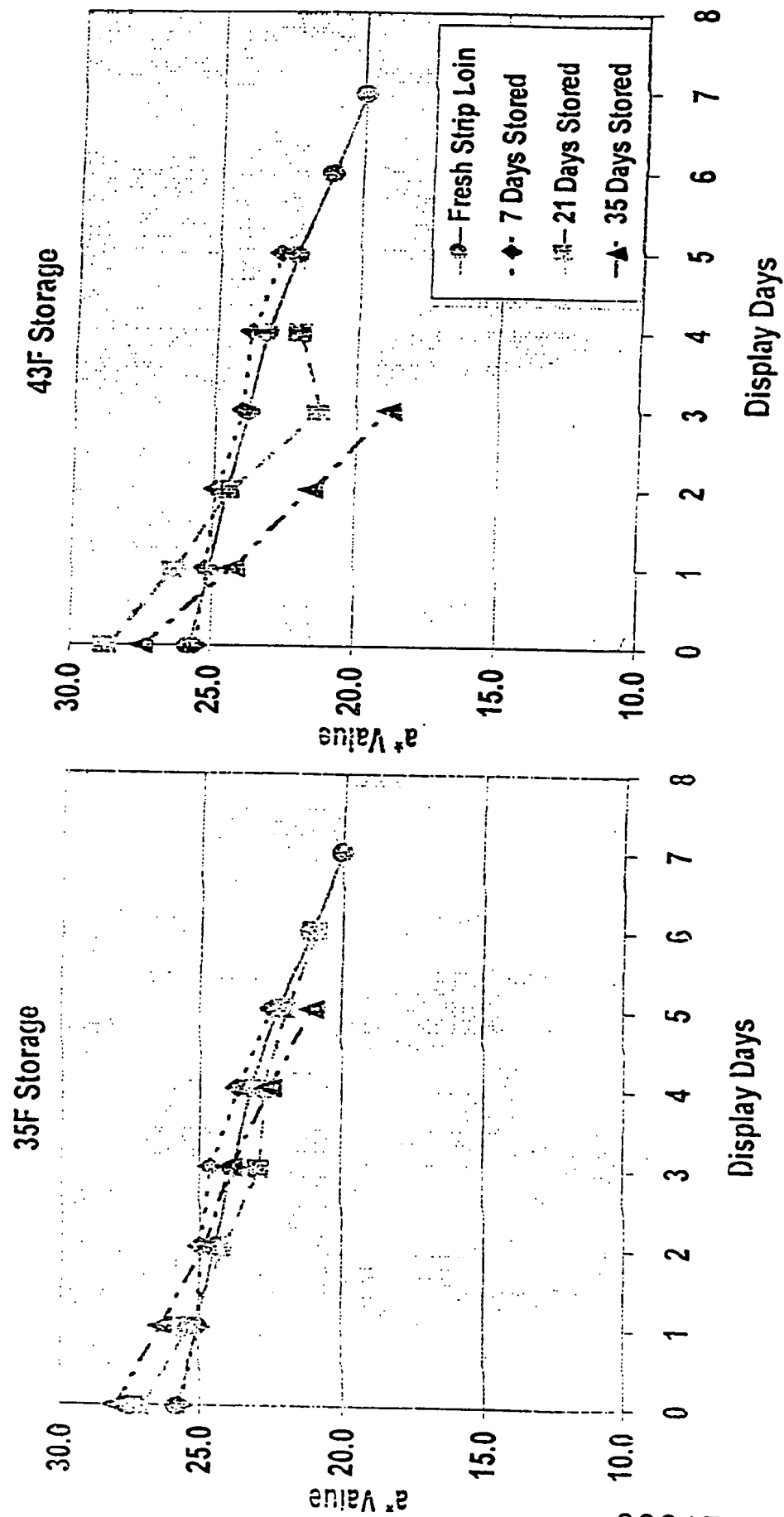


Figure 8
Inside Round (inside portion) a^* Values (Redness)
Deterioration During Display Following Storage

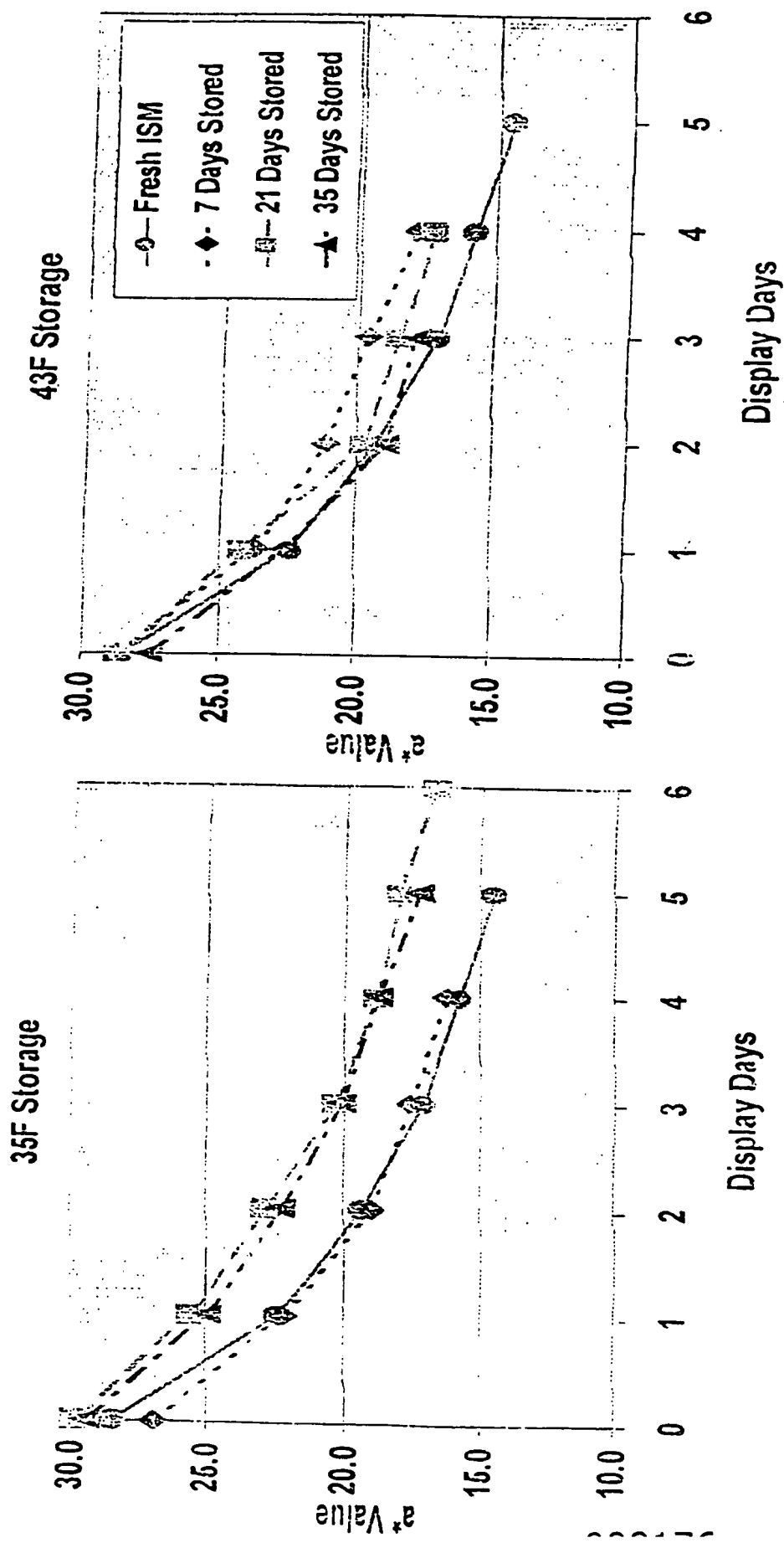


Figure 9
Inside Round (outside portion) a^* Values (Redness)
Deterioration During Display Following Storage

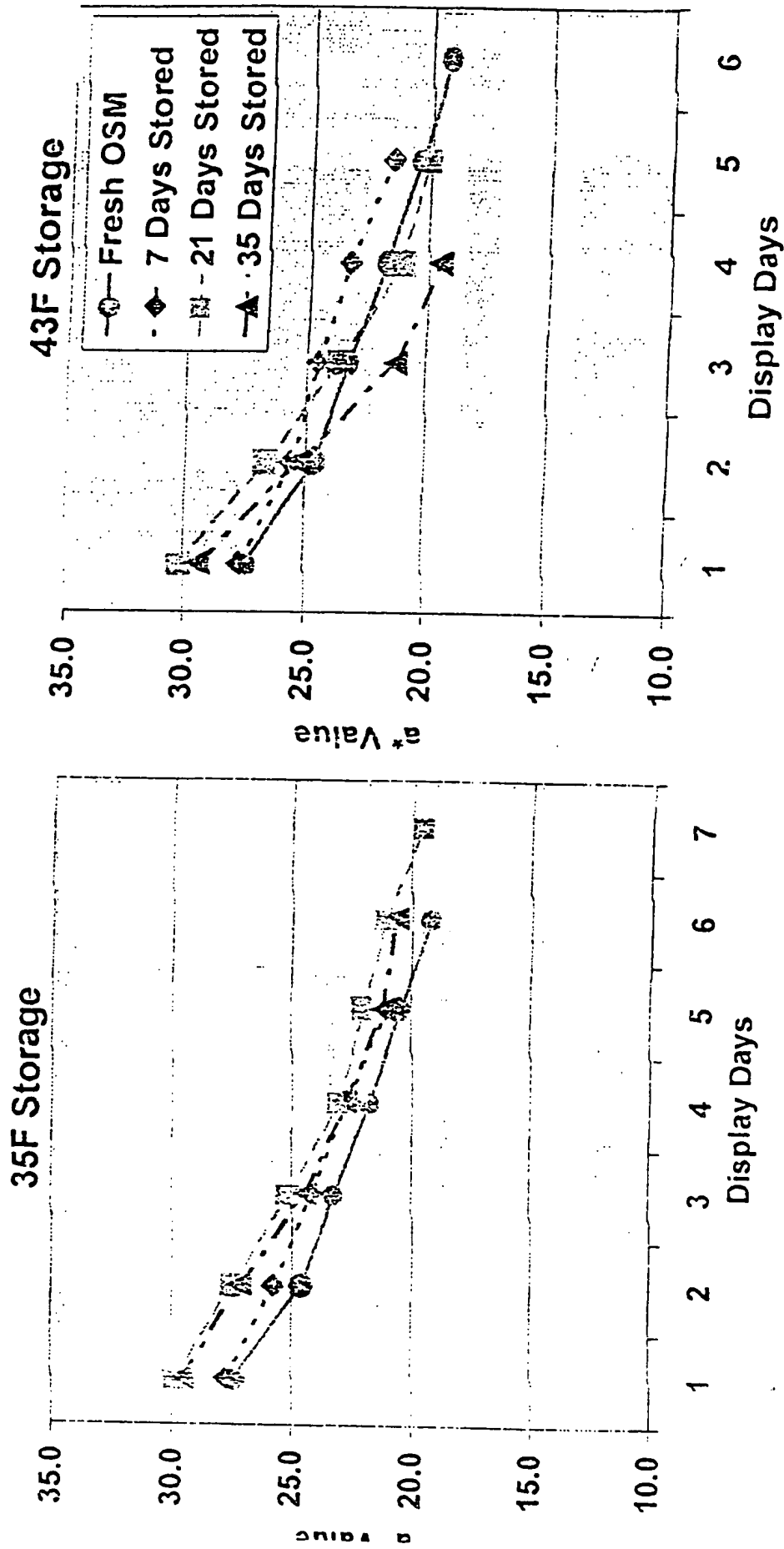


Figure 10
Tenderloin a^* Values (Redness) Deterioration
During Display Following Storage

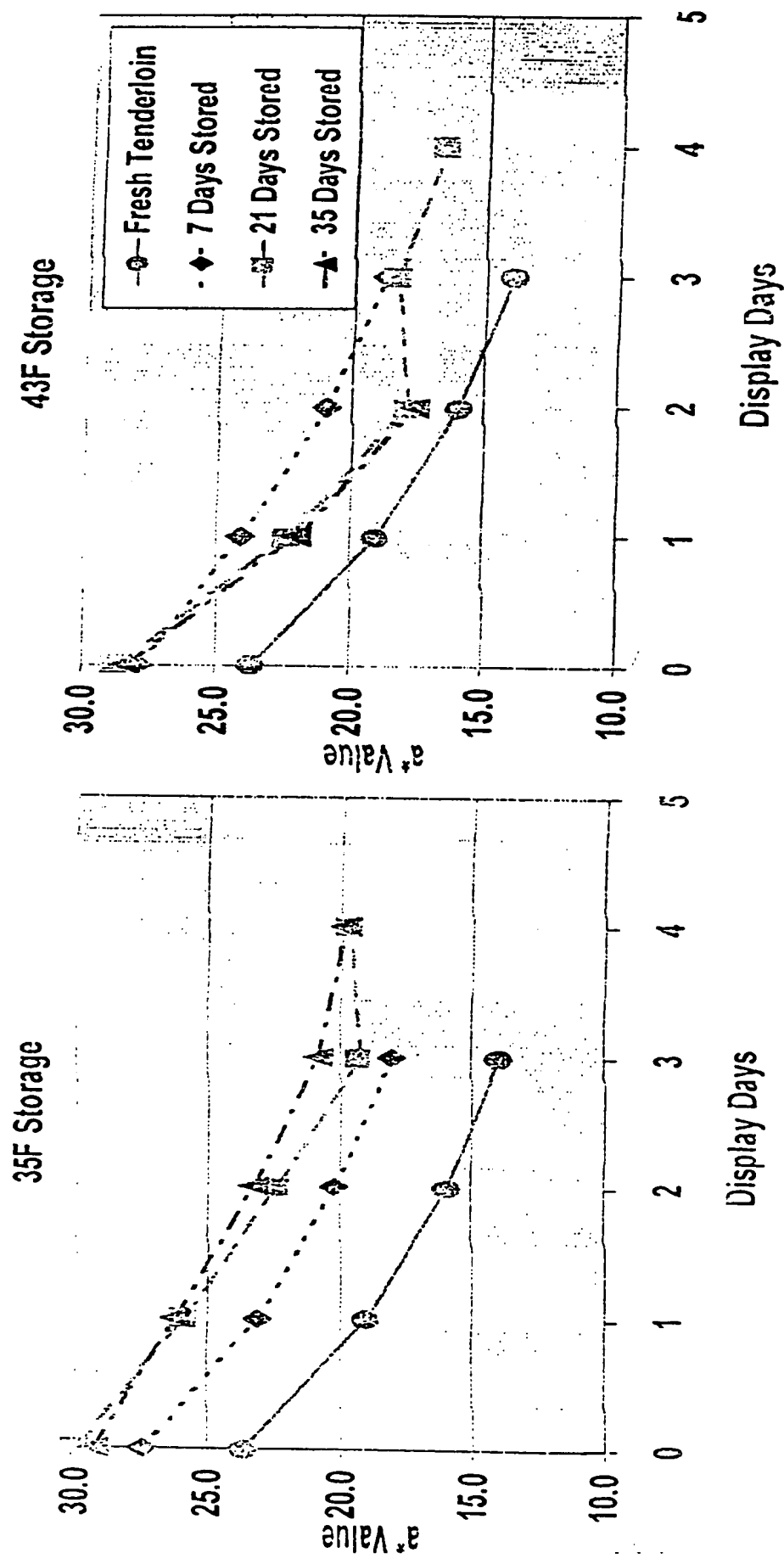


Figure 11
Ground Beef Total Aerobic Plate Counts
During Display Following Storage

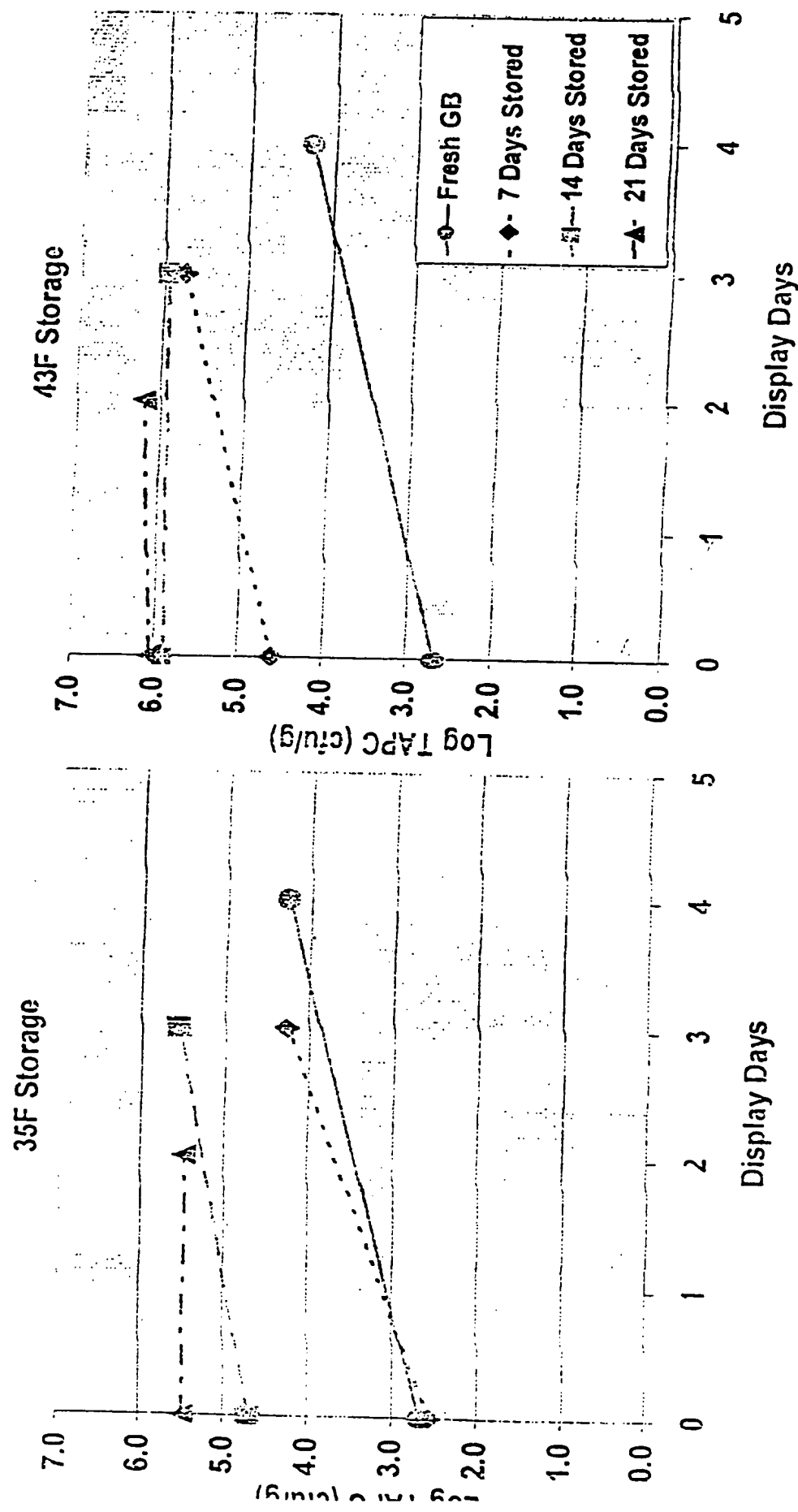


Figure 12
Strip Loin Total Aerobic Plate Counts
During Display Following Storage

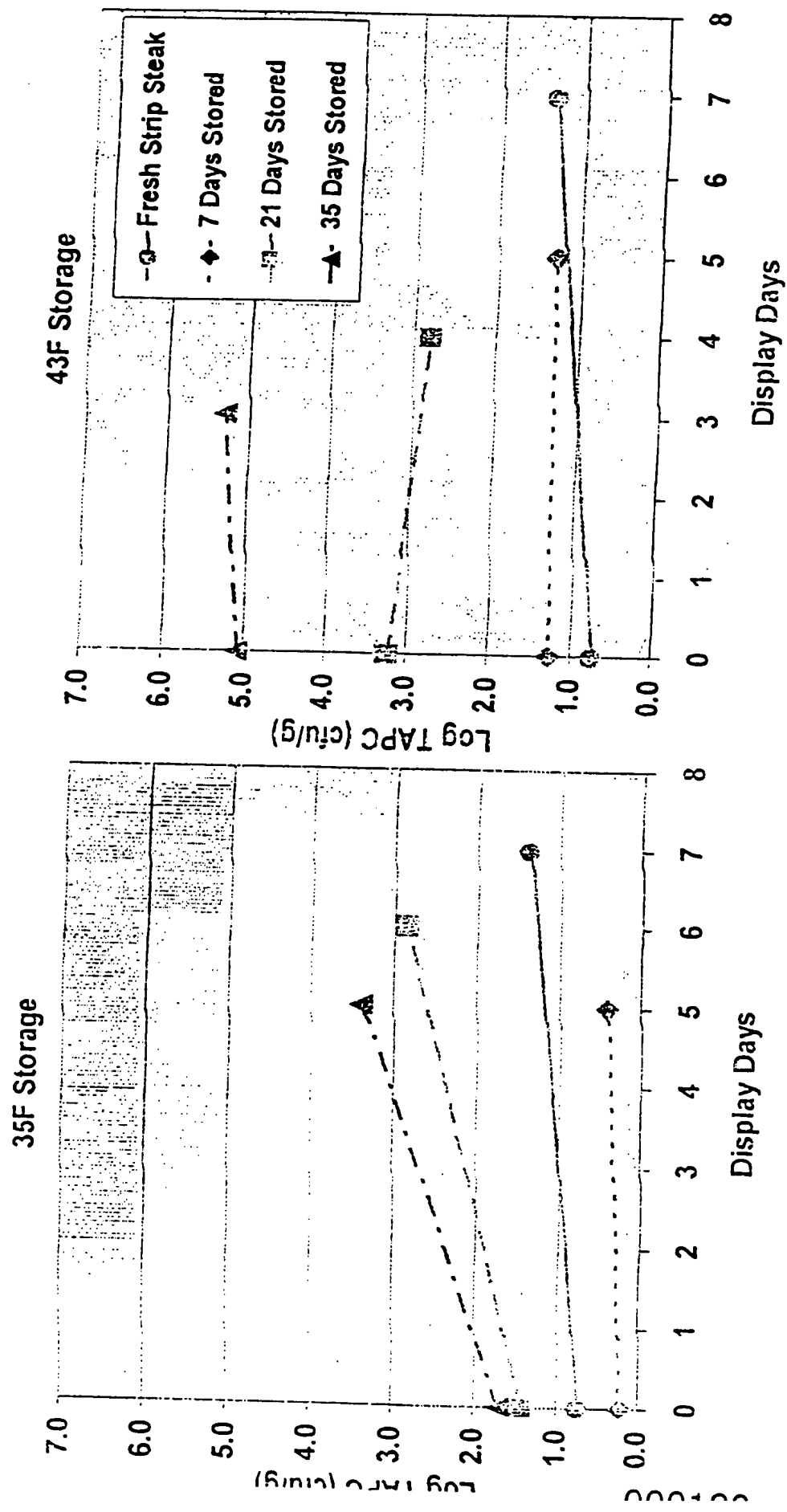


Figure 13
Inside Round Total Aerobic Plate Counts
During Display Following Storage

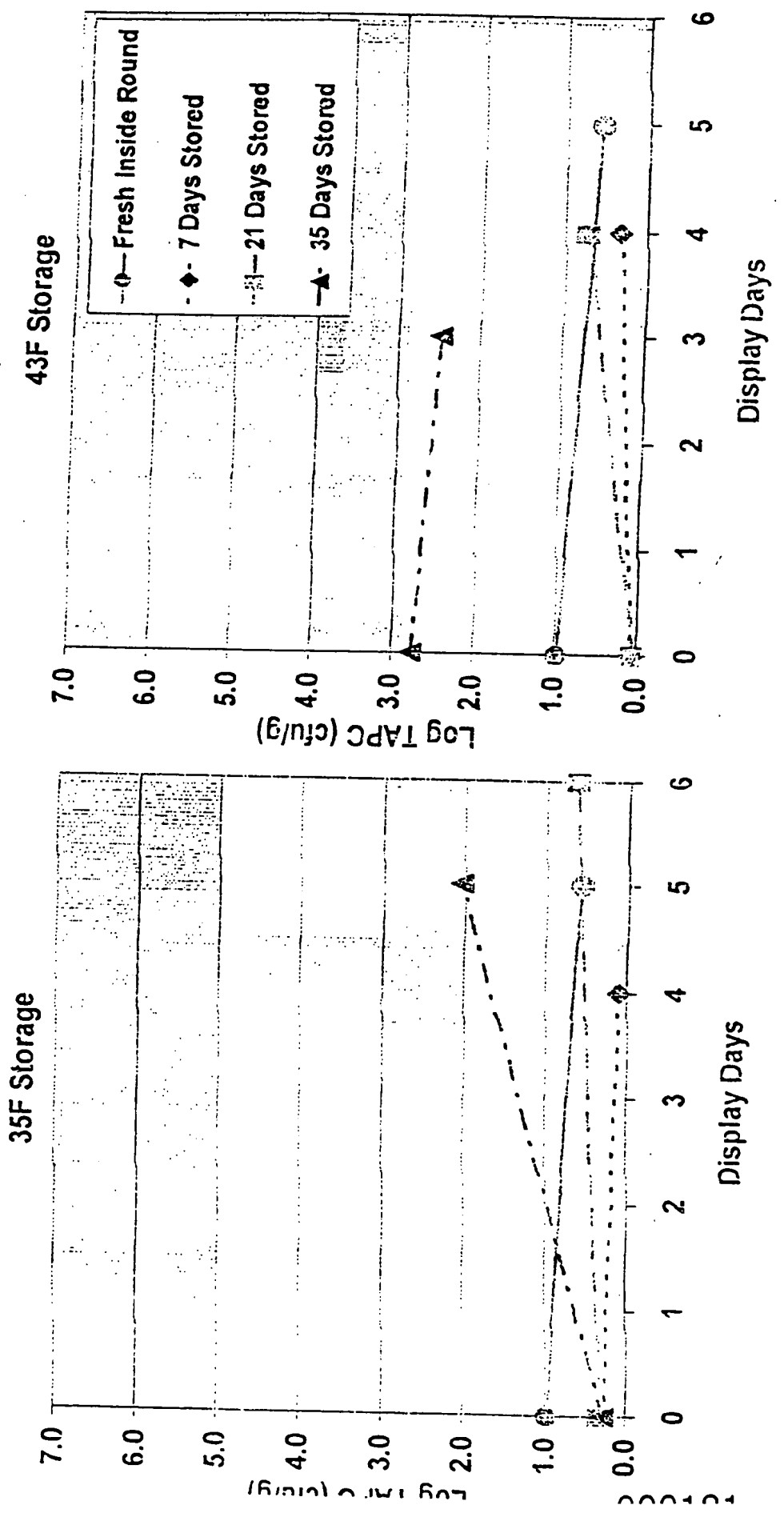


Figure 14
Tenderloin Total Aerobic Plate Counts
During Display Following Storage

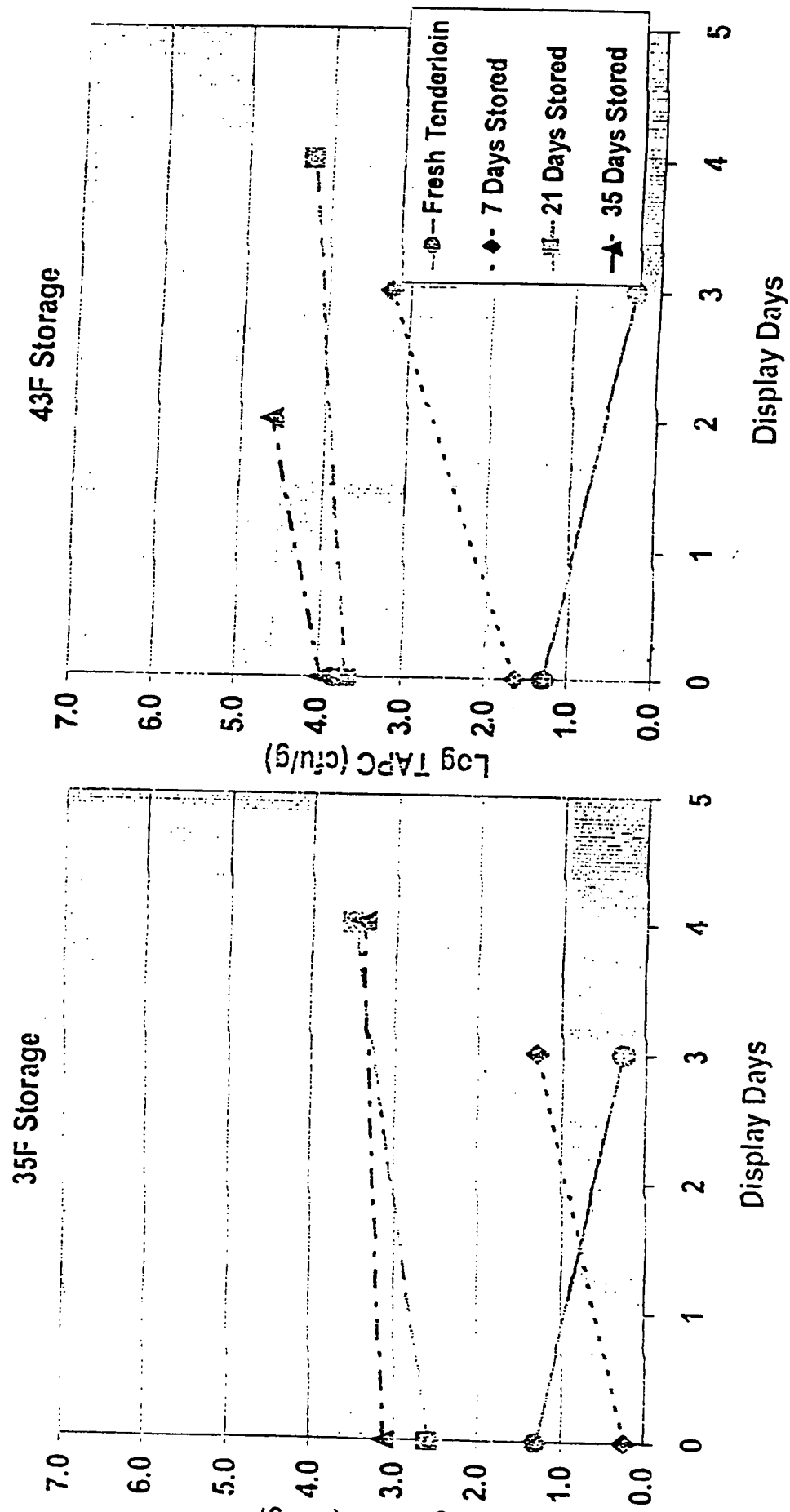


Figure 15
Ground Beef Lactic Acid Bacteria
During Display Following Storage

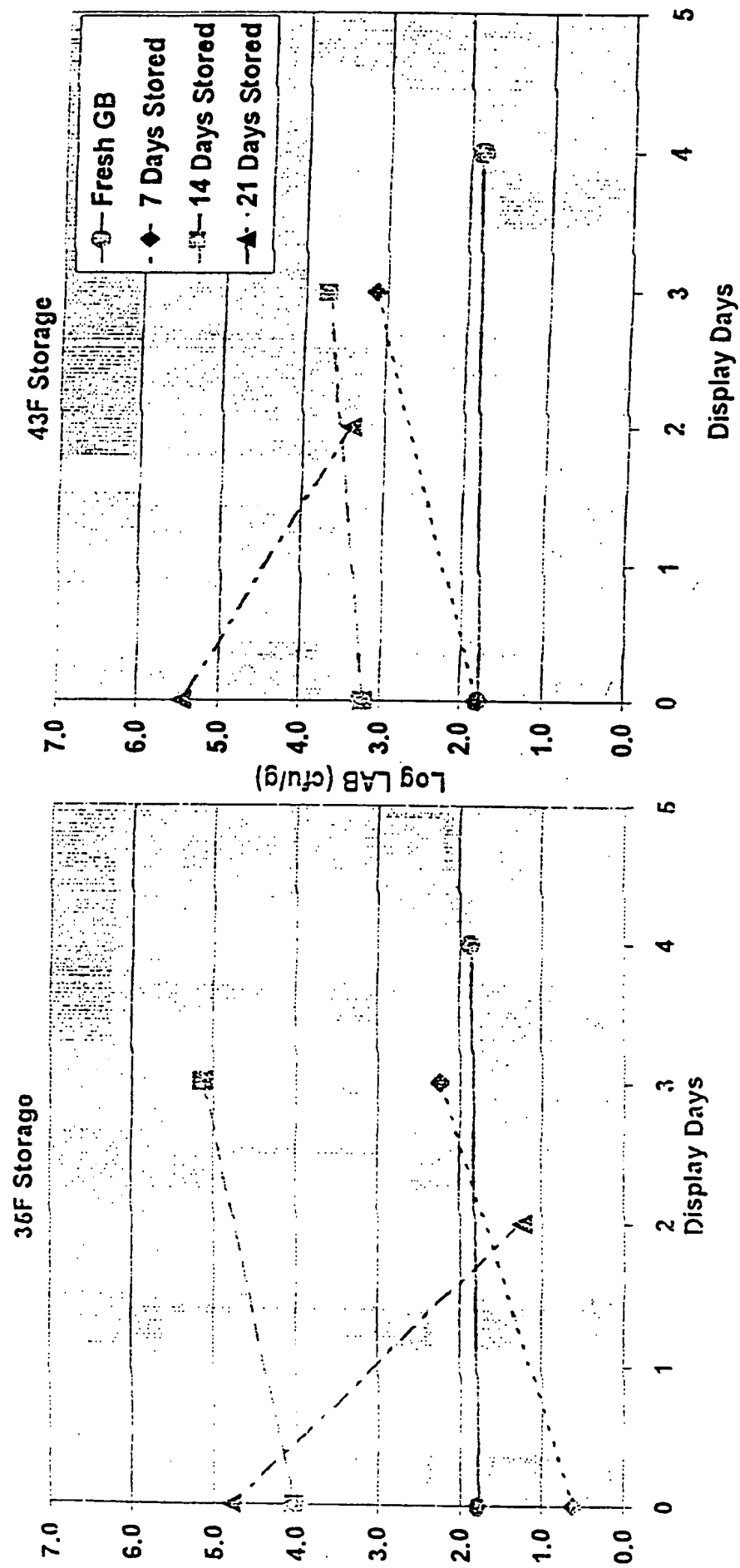


Figure 16
Strip Loin Lactic Acid Bacteria
During Display Following Storage

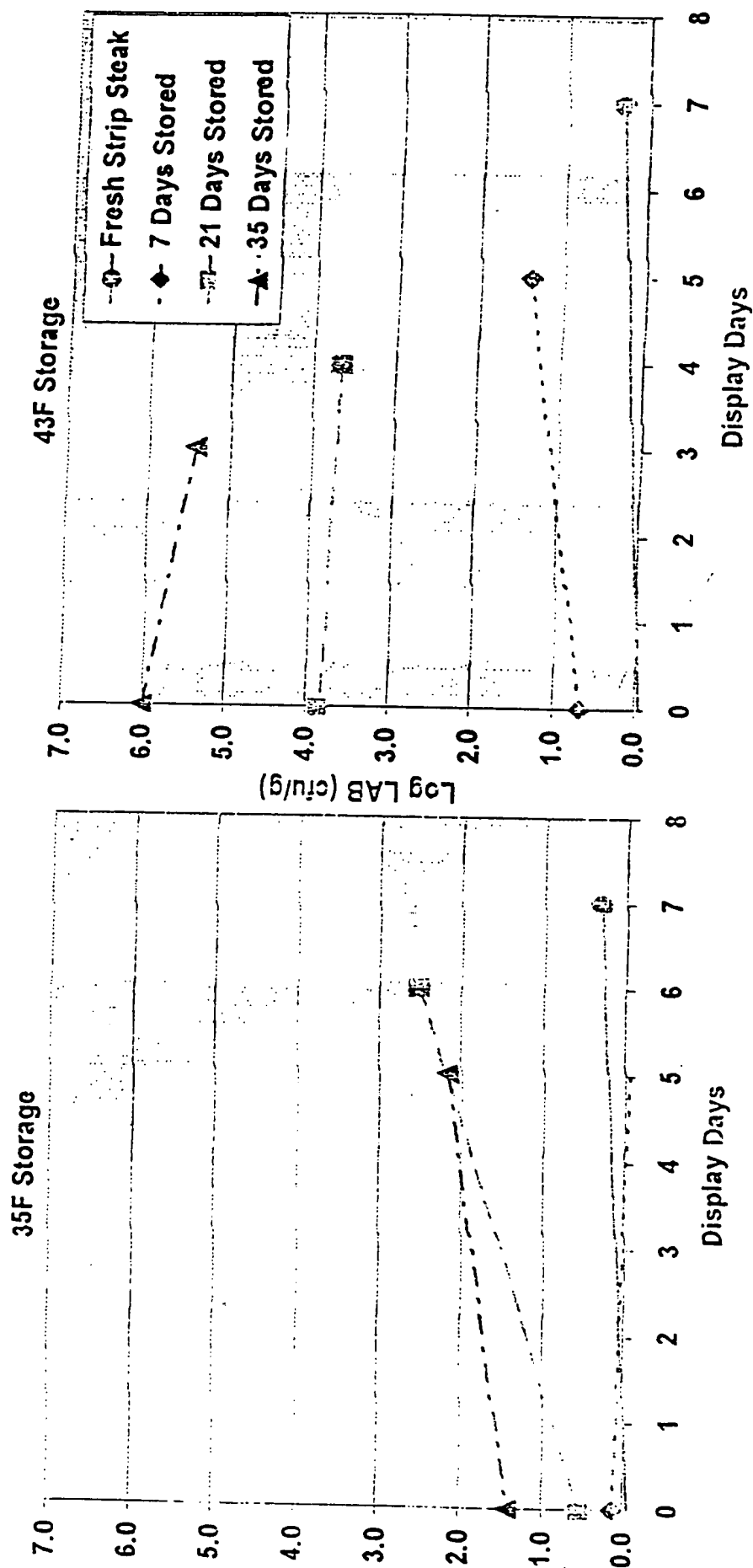


Figure 17
Inside Round Lactic Acid Bacteria
During Display Following Storage

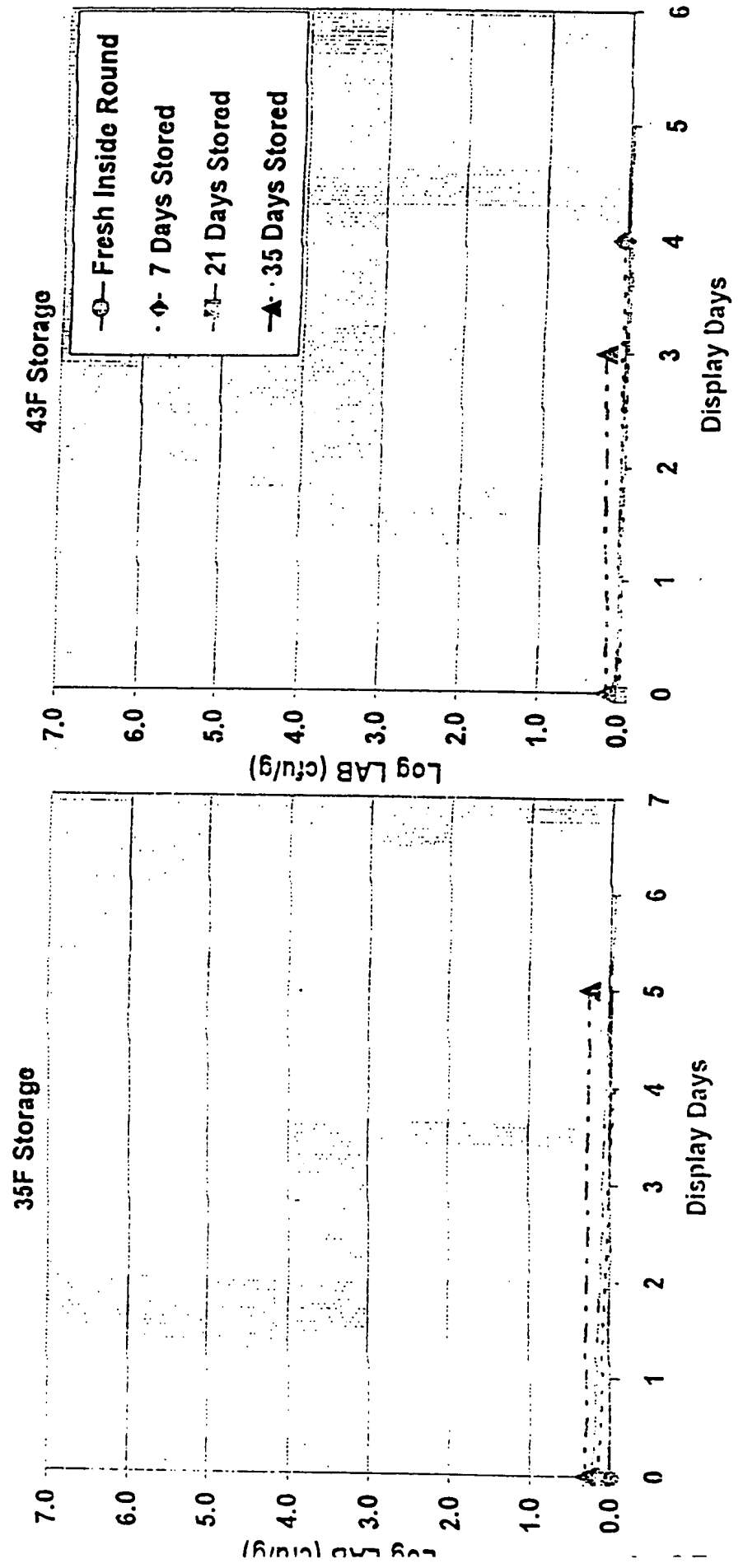


Figure 18
Tenderloin Lactic Acid Bacteria
During Display Following Storage

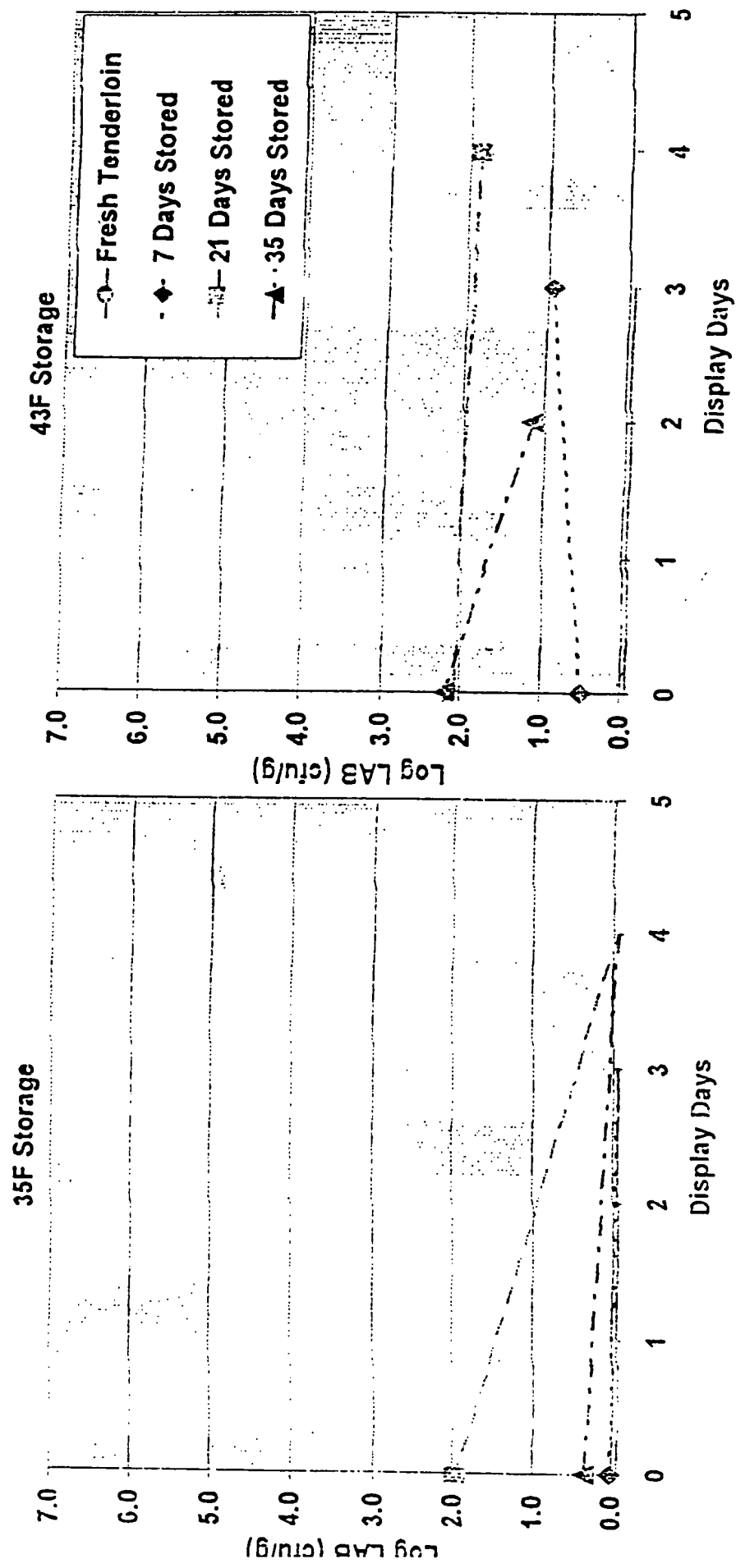


Figure 19
Aerobic Plate Count Log_{10} CFU vs Visual Color

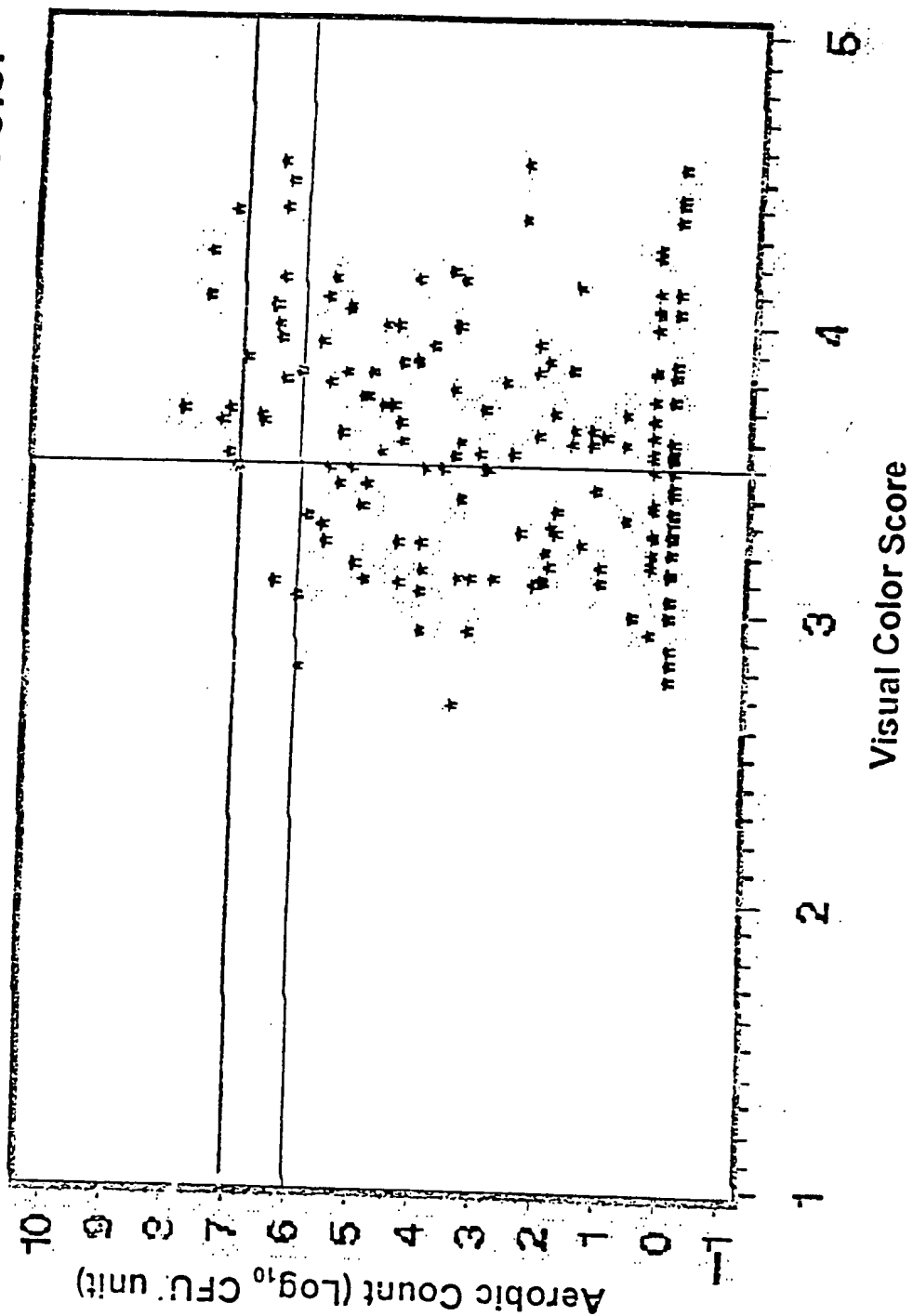


Figure 20
Lactic Acid Bacteria Count Log_{10} CFU vs Visual Color

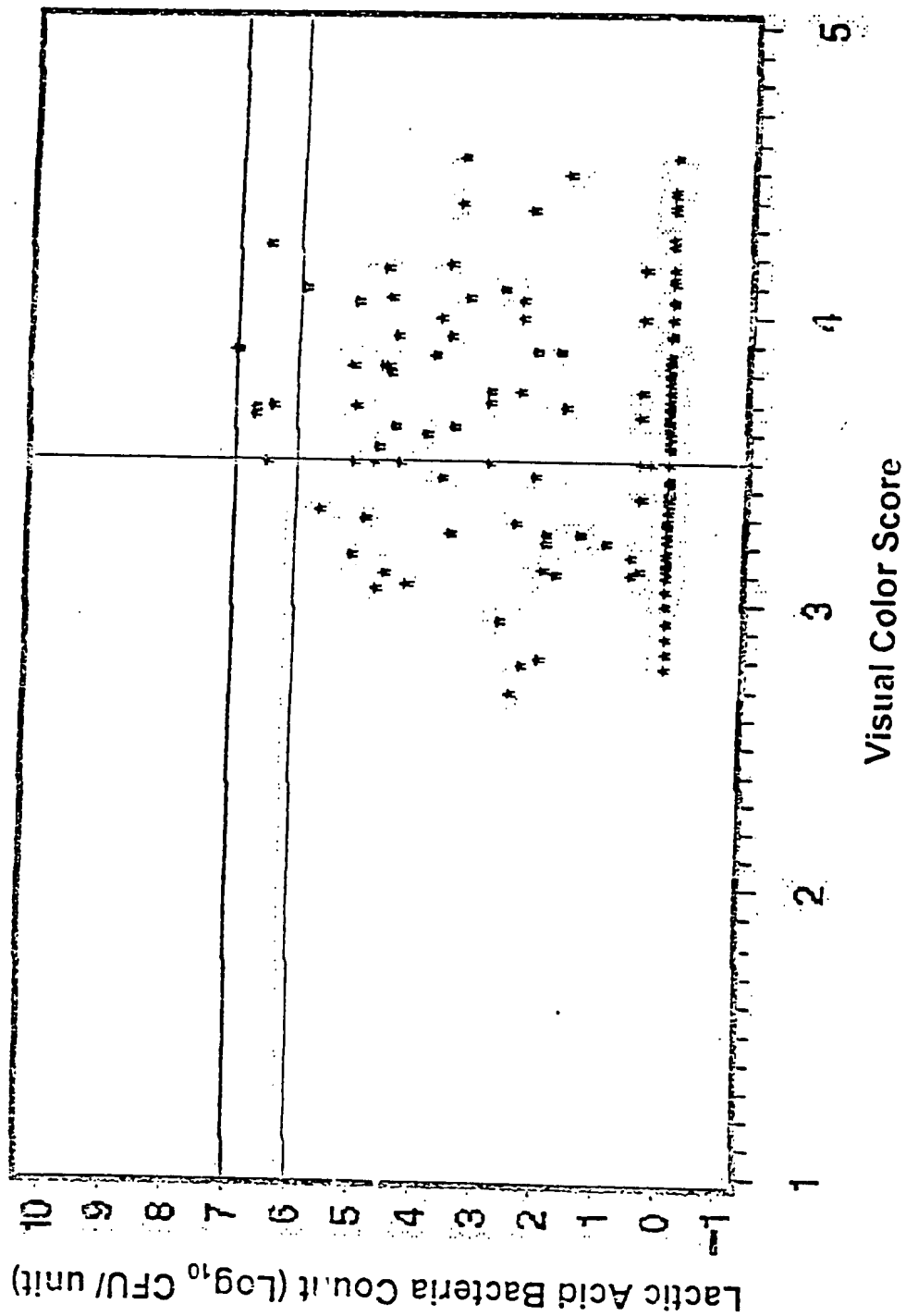
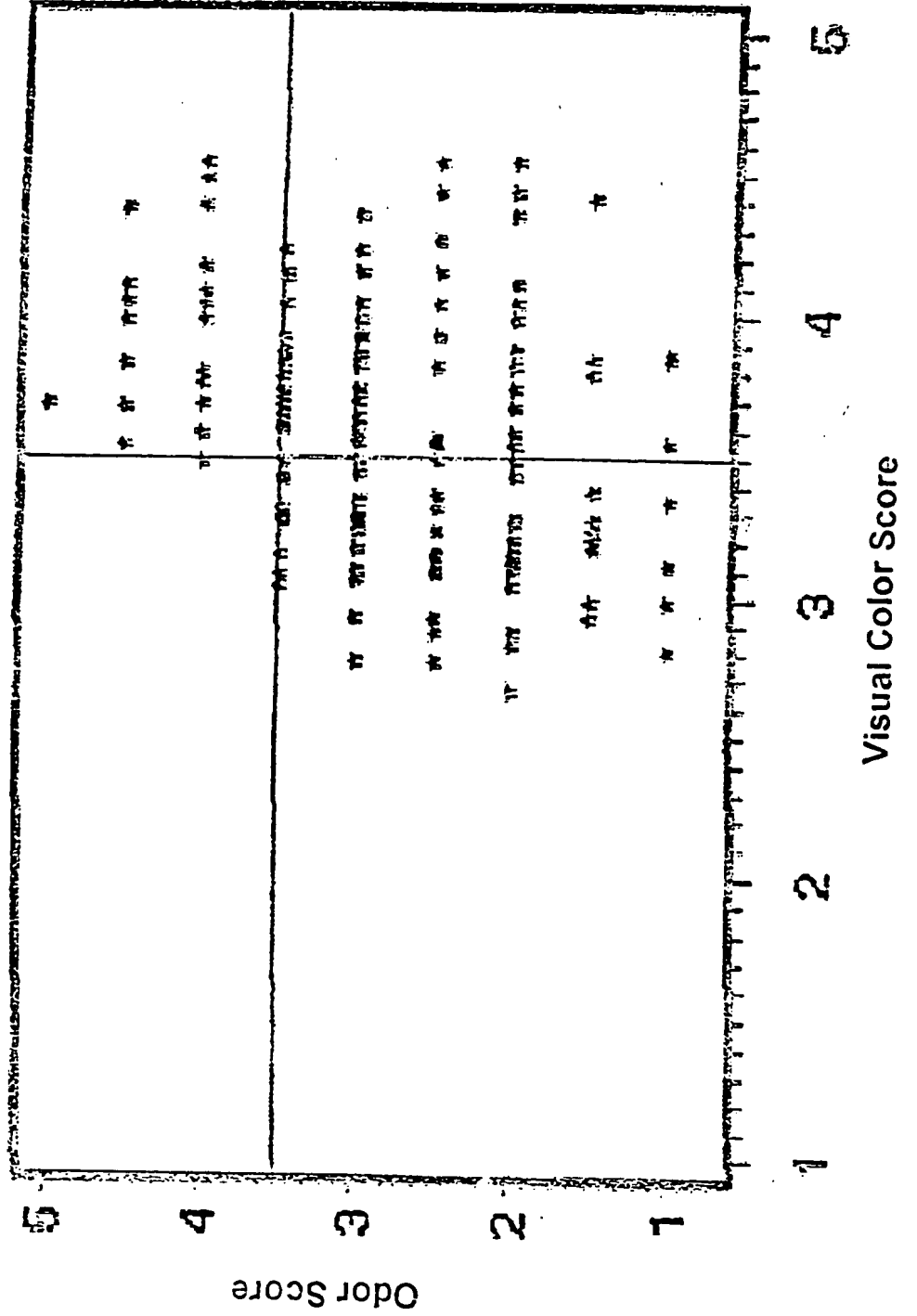


Figure 21
Odor vs. Visual Color



ATTACHMENT 5



WELDING SUPPLY, INC.

SPECIALTY GASES DIVISION

Certificate of Conformance

6000 COURT STREET ROAD
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(315) 225-3378

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FULTON, NEW YORK 13069
(315) 552-6012

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(315) 785-5438

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5 GREGORY DRIVE
SOUTH BURLINGTON, VT 05403
(802) 862-4572

48 DORAN AVENUE
GENEVA, NEW YORK 14456
(315) 781-6880

Customer: Pactiv Packaging

Material Submitted: Carbon Monoxide, Research Purity 99.99% min.

Date Reported: 05/08/01

Component	Specification
Carbon Monoxide	99.99% min.
Oxygen	< 0.5 PPM
Nitrogen	< 10 PPM
Carbon Dioxide	< 20 PPM
Methane	< 5 PPM
Ethane	< 1 PPM
Propane	< 1 PPM
Dimethyl Ether	< 1 PPM
Hydrogen	< 1 PPM
Moisture	< 1 PPM

Note: Analysis are conducted utilizing approved analytical method (s) and are correct to within the analytical accuracies of this (these) method (s).

Quality Control Approved

Lorrie Nash

05-08-01

ATTACHMENT 6



Standard Practice for Analysis of Reformed Gas by Gas Chromatography¹

This standard is issued under the fixed designation D 1946; the number immediately following the designation indicates the year of original adoption or, in the case of revision, the year of last revision. A number in parentheses indicates the year of last reapproval. A superscript epsilon (ε) indicates an editorial change since the last revision or reapproval.

1. Scope

1.1 This practice covers the determination of the chemical composition of reformed gases and similar gaseous mixtures containing the following components: hydrogen, oxygen, nitrogen, carbon monoxide, carbon dioxide, methane, ethane, and ethylene.

1.2 *This standard does not purport to address all of the safety concerns, if any, associated with its use. It is the responsibility of the user of this standard to establish appropriate safety and health practices and determine the applicability of regulatory limitations prior to use.*

2. Referenced Documents

2.1 ASTM Standards:

E 260 Practice for Packed Column Gas Chromatography²

3. Summary of Practice

3.1 Components in a sample of reformed gas are physically separated by gas chromatography and compared to corresponding components of a reference standard separated under identical operating conditions, using a reference standard mixture of known composition. The composition of the reformed gas is calculated by comparison of either the peak height or area response of each component with the corresponding value of that component in the reference standard.

4. Significance and Use

4.1 The information about the chemical composition can be used to calculate physical properties of the gas, such as heating (calorific) value and relative density. Combustion characteristics, products of combustion, toxicity, and interchangeability with other fuel gases may also be inferred from the chemical composition.

5. Apparatus

5.1 **Detector**—The detector shall be a thermal conductivity type or its equivalent in stability and sensitivity. The thermal conductivity detector must be sufficiently sensitive to produce

a signal of at least 0.5 mV for 1 mol % methane in a 0.5-mL sample.

5.2 **Recording Instruments**—Either strip chart recorders or electronic integrators, or both, are used to display the separated components. Although a strip chart recorder is not required when using electronic integration, it is highly desirable for evaluation of instrument performance.

5.2.1 The recorder, when used, shall be a strip chart recorder with a full-range scale of 5 mV or less (1 mV preferred). The width of the chart shall be not less than 150 mm. A maximum pen response time of 2 s (1 s preferred) and a minimum chart speed of 10 mm/min shall be required. Faster speeds up to 100 mm/min are desirable if the chromatogram is to be interpreted using manual methods to obtain areas.

5.2.2 **Electronic or Computing Integrators**—Proof of separation and response equivalent to that for the recorder is required for displays other than by chart recorder.

5.3 **Attenuator**—If manual methods are used to interpret the chromatogram, an attenuator must be used with the detector output signal to keep the peak maxima within the range of the recorder chart. The attenuator must be accurate to within 0.5 % between the attenuator range steps.

5.4 Sample Inlet System:

5.4.1 The sample inlet system must be constructed of materials that are inert and nonadsorptive with respect to the components in the sample. The preferred material of construction is stainless steel. Copper and copper-bearing alloys are unacceptable.

5.4.2 Provision must be made to introduce into the carrier gas ahead of the analyzing column a gas-phase sample that has been entrapped in either a fixed volume loop or tubular section. The injected volume must be reproducible such that successive runs of the same sample agree within the limits of repeatability for the concentration range as specified in 11.1.1.

5.4.3 If the instrument is calibrated with pure components, the inlet system shall be equipped to introduce a sample at less than atmospheric pressure. The pressure-sensing device must be accurate to 0.1 kPa (1 mm Hg).

5.5 Column Temperature Control:

5.5.1 **Isothermal**—When isothermal operation is used, the analytical columns shall be maintained at a temperature constant to 0.3°C during the course of the sample run and the corresponding reference run.

¹ This practice is under the jurisdiction of ASTM Committee D-3 on Gaseous Fuels and is the direct responsibility of Subcommittee D03.07 on Analysis of Chemical Composition of Gaseous Fuels.

Current edition approved March 30, 1990. Published May 1990. Originally published as 1946 – 62 T. Last previous edition D 1946 – 82.

² Annual Book of ASTM Standards, Vol 14.02.

5.5.2 Temperature Programming—Temperature programming may be used, as feasible. The oven temperature shall not exceed the recommended temperature limit for the materials in the column.

5.6 Detector Temperature Control—The detector temperature shall be maintained at a temperature constant to 0.3°C during the course of the sample run and the corresponding reference run. The detector temperature shall be equal to, or greater than, the maximum column temperature.

5.7 Carrier Gas—The instrument shall be equipped with suitable facilities to provide flow of carrier gas through the analyzer and detector at a flow rate that is constant to 1 % throughout the analysis of the sample and the reference standard. The purity of the carrier gas may be improved by flowing the carrier gas through selective filters before its entry into the chromatograph.

5.8 Columns:

5.8.1 The columns shall be constructed of materials that are inert and nonadsorptive with respect to the components in the sample. The preferred material of construction is stainless steel. Copper and copper-bearing alloys are unacceptable.

5.8.2 Either an adsorption-type column or a partition-type column, or both, may be used to make the analysis.

NOTE 1—See Practice E 260 for general gas chromatography procedures.

5.8.2.1 Adsorption Column—This column must completely separate hydrogen, oxygen, nitrogen, methane, and carbon monoxide. If a recorder is used, the recorder pen must return to the baseline between each successive peak. Equivalent proof of separation is required for displays other than by chart recorder. Fig. 1 is an example chromatogram obtained with an adsorp-

tion column.

(1) Because of similarities in thermal conductivities, helium should not be used as the carrier gas for hydrogen when hydrogen is less than 1 % of the sample. Either argon or nitrogen carrier gas is suitable for both percent and parts per million quantities of hydrogen.

(2) The use of a carrier gas mixture of 8.5 % hydrogen and 91.5 % helium will avoid the problem of reversing polarities of hydrogen responses as the concentration of hydrogen in the sample is increased.

(3) The precision of measurement of hydrogen can be increased by using a separate injection for hydrogen, using either argon or nitrogen for the carrier gas.

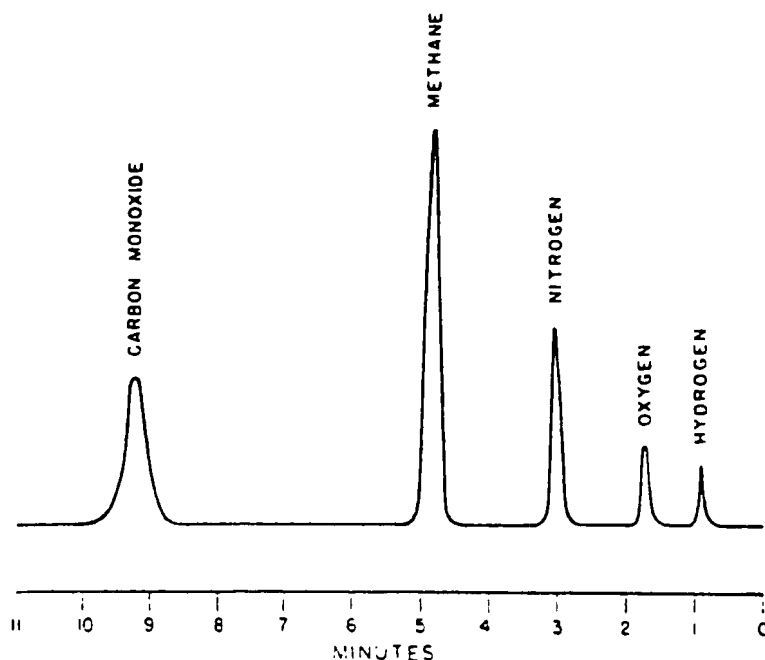
(4) Another technique for isolating the hydrogen in a sample is to use a palladium transfer tube at the end of the adsorption column; this will permit only hydrogen to be transferred to a stream of argon or nitrogen carrier gas for analysis in a second thermal conductivity detector.

5.8.2.2 Partition Column—This column must separate ethane, carbon dioxide, and ethylene. If a recorder is used, the recorder pen must return to the baseline between each successive peak. Equivalent proof of separation is required for displays other than by chart recorder. Fig. 2 is an example chromatogram obtained with a partition column.

5.8.3 General—Those column materials, operated either isothermally or with temperature programming, or both, may be used if they provide satisfactory separation of components.

6. Reference Standards

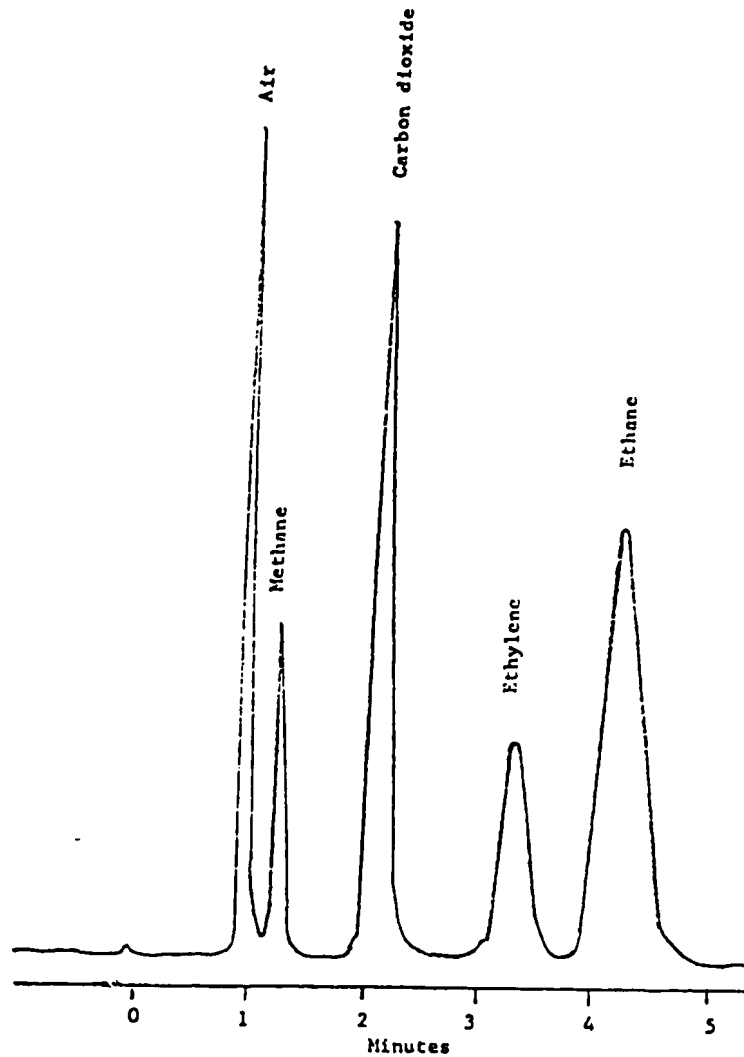
6.1 Moisture-free mixtures of known composition are required for comparison with the test sample. They must contain known percentages of the components, except oxygen (Note



Column: 2-m by 6-mm inside diameter Type 13 x molecular sieves, 14 to 30 mesh
Temperature: 35°C

Flow rate: 60-mL helium/min
Sample size: 0.5 mL

FIG. 1 Chromatogram of Reformed Gas on Molecular Sieve Column



Column: 1.2 m by 6.35 mm
 Porapak Q, 50 to 80 mesh
 Current setting: 225 mA

Temperature: 40°C
 Flow rate: 50-mL helium/min
 Sample size: 0.5 mL

FIG. 2 Chromatogram of Reformed Gas on Porapak Q Column

2), that are to be determined in the unknown sample. All components in the reference standard must be homogeneous in the vapor state at the time of use. The fraction of a component in the reference standard should not be less than one half of, nor differ by more than 10 mol % from, the fraction of the corresponding component in the unknown. The composition of the reference standard must be known to within 0.01 mol % for any component.

NOTE 2—Unless the reference standard is stored in a container that has been tested and proved for inertness to oxygen, it is preferable to calibrate for oxygen by an alternative method.

6.2 *Preparation*—A reference standard may be prepared by blending pure components. Diluted dry air is a suitable standard for oxygen and nitrogen.

NOTE 3—A mixture containing approximately 1 % of oxygen can be prepared by pressurizing a container of dry air at atmospheric pressure to 20 atm (2.03 MPa) with pure helium. This pressure need not be measured

precisely, as the fraction of nitrogen in the mixture such prepared must be determined by comparison to nitrogen in the reference standard. The fraction of nitrogen is multiplied by 0.280 to obtain the fraction of oxygen plus argon. Argon elutes with oxygen in the molecular sieves column. Do not rely on oxygen standards that have been prepared for more than a few days. It is permissible to use a response factor for oxygen that is relative to a stable component.

7. Preparation of Apparatus

7.1 *Column Preparation*—Pack a 2- to 3-m column (6-mm inside diameter stainless steel tubing) with Type 13X molecular sieves, 14 to 30 mesh, that have been dried 12 h or more at 300 to 350°C. Pack a second column (1 m by 6 mm) with Porapak Q,³ 50 to 80 mesh, that has been dried 12 h or more at about 150°C. Shape the columns to fit the configuration of the oven in the chromatograph.

³ Available from Waters Associates, Inc., Framingham, MA 01701.

NOTE 4—Variations in column material, dimensions, and mesh sizes of packing are permissible if the columns produce separations equivalent to those shown in Fig. 1 and Fig. 2. Better performance may be obtained by using a 2.1-mm stainless steel tubing with corresponding smaller mesh packing materials and substituting Haysep Q for Porapak Q.

7.2 Chromatograph—Place the proper column and sample volume in operation for the desired run in accordance with 8.1 and 8.2. For isothermal operation, the column should be maintained at a temperature between 30 and 45°C. When appropriate, column temperatures may be increased. Adjust the operating conditions and allow the instrument to stabilize. Check the stability by making repeat runs on the reference standard to obtain reproducible peak heights as described in 5.4.2 for corresponding components.

8. Procedure

8.1 Sample Volume—The sample introduced into the chromatographic column should have a volume between 0.2 and 0.5 mL. Sufficient accuracy can be obtained for the determination of all but the very minor components with this sample size. When increased sensitivity is required for the determination of components present in low concentrations, a sample size of up to 5 mL is permissible. However, components whose concentrations are in excess of 5 % should not be analyzed by using sample volumes greater than 0.5 mL.

8.2 Chromatograms:

8.2.1 Adsorption Column (Fig. 1)—Obtain a steady baseline on the recorder with a constant carrier gas flowrate appropriate to the column diameter. Introduce a sample of the unknown mixture at atmospheric pressure into the chromatograph and obtain a response similar to that of Fig. 1 of the components hydrogen, oxygen, nitrogen, methane, and carbon monoxide, which elute in that order. Repeat with a sample of the reference standard. If oxygen is present in the mixture, run a sample of air, either at an accurately measured reduced pressure, or air freshly diluted with helium, so that the partial pressure of oxygen is approximately equal to that of the oxygen in the mixture being analyzed.

NOTE 5—The peak for carbon monoxide can appear between those of nitrogen and methane if the molecular sieves have become contaminated. If this occurs, replace or regenerate the column packing by heating in accordance with 7.1.

8.2.2 Partition Column (Fig. 2)—Establish a steady baseline with the helium carrier gas flowing through the Porapak Q column. Introduce a sample of the reference standard and then a sample of the unknown mixture. Obtain responses similar to that shown in Fig. 2 for carbon dioxide, ethane, and ethylene.

8.2.3 All chromatograms for manual measurement should be run at a sensitivity setting that permits maximum peak height to be recorded for each component.

8.2.4 Column isolation valves may be used to make the entire analysis with a single injection if the separations specified in 5.8.2.1 and 5.8.2.2 are produced.

9. Calculation

9.1 The number of significant digits retained for the quantitative value of each component shall be such that accuracy is neither sacrificed nor exaggerated. The expressed numerical

value of any component in the sample should not be presumed to be more accurate than the corresponding certified value of that component in the calibration standard.

9.2 Manual Measurement—Measure the response of each component, convert to the same sensitivity for corresponding components in the sample and reference standard, and calculate the mole percent of each component in the sample as follows:

$$C = (A/B)(S) \quad (1)$$

where:

C = mole percent of the component in the sample,

A = response of the component in the sample,

B = response of the component in the standard at the same sensitivity as with A , and

S = mole percent of the component in the reference standard.

9.3 If a helium-diluted air mixture was run for oxygen calibration, calculate the fraction of oxygen in the mixture from the fraction of the nitrogen and the composition of the diluted air. Calculate the fraction of nitrogen in the mixture in accordance with 9.1, using the nitrogen response of the reference standard for comparison. Air composition values of 78.1 % nitrogen and 21.9 % oxygen should be used, as argon (0.9 % in air) elutes with oxygen on the molecular sieves column.

9.4 If air has been analyzed at reduced pressure to calibrate for oxygen, correct the equation for pressure as follows:

$$C = (A/B)(S)(P_a/P_b) \quad (2)$$

where:

P_a = absolute pressure at which air was analyzed and

P_b = barometric pressure when sample was analyzed, with both pressures being expressed in the same units.

9.5 Normalize the mole percent values by multiplying each value by 100 and dividing by the sum of the original values. The sum of the original values should not differ from 100.0 % by more than 1.0 %.

10. Analysis of the Reference Standard

10.1 If the composition of the reference standard is not known to a sufficient degree of accuracy, analyze it by the use of pure components for calibration. Obtain chromatograms of the standard as described in 8.2, except measure the pressure of each sample introduced to 0.133 kPa (1 mm Hg). When each chromatogram is obtained, calibrate each component by introducing a sample of the pure component at a pressure that closely approximates its partial pressure in the blend (for example, a component whose concentration in the standard is 50 % is analyzed at 50 % of the pressure at which the standard was analyzed). Use a minimum pressure of 0.665 kPa (5 mm Hg) for minor components. Repeat the analysis with the reference standard. Corresponding peak heights should agree within 1 mm or 1 % (whichever is larger) when recorded on a sensitivity setting that allows maximum response on the recorder chart.

10.2 Calculate the composition of the reference standard by the adjustment of responses of like components to the same sensitivity and calculate the concentration of each component as follows:

$$C = \frac{(100)(R)(P_p)}{(P)(P_r)} \quad (3)$$

where:

- C = component concentration, mole percent;
- R = response of the component in the reference standard;
- P = response of the pure component;
- P_p = pressure at which the pure component was analyzed; and
- P_r = pressure at which the reference standard was analyzed, with both pressures being expressed in the same absolute units.

10.2.1 Normalize all values as described in 9.4.

11. Precision

11.1 The following data should be used to judge the acceptability of the results:

11.1.1 *Repeatability*—Duplicate results by the same operator should not be considered suspect unless they differ by more

than the following amounts:

Component, mol %	Repeatability
0 to 1	0.05
1 to 5	0.1
5 to 25	0.3
Over 25	0.5

11.1.2 *Reproducibility*—Results submitted by different laboratories should not differ by more than the amounts given in 11.1.1 when the same reference standard is used for calibration and the same composition is used for calculations. If calibration is made with pure components or with different reference standards, results submitted by each of two laboratories should not be considered suspect unless the results differ by more than the following amounts:

Component, mol %	Reproducibility
0 to 1	0.1
1 to 5	0.2
5 to 25	0.5
Over 25	1.0

12. Keywords

12.1 gaseous fuels

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ATTACHMENT 7

Utah State UNIVERSITY

DEPARTMENT OF NUTRITION AND FOOD SCIENCES
College of Agriculture
College of Family Life
Logan, UT 84322-8700
Telephone: (435) 797-2126
FAX: (435) 797-2379

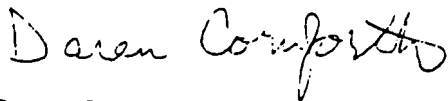
Division of GRAS Notice Review
Office of Food Additive Safety
Center for Food Safety and Applied Nutrition
Food and Drug Administration
200 C St, SW
Washington, DC 20204

August 17, 2001

Dear FDA Personnel,

I am a meat scientist with experience in studying the effects on fresh meats of various modified atmospheres. Based on my review of the details of the ActiveTech 2001 modified atmosphere system employing 0.4% carbon monoxide gas in a mixture with 60 percent carbon dioxide and the remainder nitrogen, as well as the published literature and common knowledge in the field, I confirm that the use of modified atmospheres including 0.4% CO to package fresh meats as used in the ActiveTech 2001 system is both safe and generally recognized as safe. If I can provide any further information or clarification, please contact me.

Sincerely yours,



Daren Cornforth, Ph.D.
Professor, Nutrition & Food Sciences
Utah State University
435-797-2114
darenc@cc.usu.edu

000199

Daren Cornforth, Ph.D.

Birthdate: May 23, 1949

Birthplace: Fort Collins, Colorado

Current Address: Department of Nutrition and Food Sciences
Utah State University
Logan, Utah 84322-8700
(435) 797-2114 fax (435) 797-2379
e-mail darenc@cc.usu.edu

Education

<u>Institution</u>	<u>Degree</u>	<u>Year Conferred</u>	<u>Scientific Field</u>
Colorado State University	B.S.	1971	Animal Science
Colorado State University	M.S.	1973	Animal Science
Michigan State University	Ph.D.	1978	Food Science & Human Nutrition

Research and Professional Experience

<u>Dates</u>	<u>Position</u>	<u>Duties</u>
1978 to present	Asst., Assoc., Full Prof., NFS	Teaching/Research, Meat Processing
1974 to 1978	Graduate Research Assistant at Michigan State University East Lansing, Michigan	Dissertation on cold- induced shortening and toughening of beef muscle.
1971 to 1973	Graduate Research Assistant at Colorado State University Fort Collins, Colorado	Thesis on the effects of breed, sex, and diet on muscle fiber type and fat cell development in growing calves.

Memberships

Institute of Food Technologists, Amer. Assoc. for the Advancement of Science, Sigma Xi (Past President, USU Chapter), American Meat Science Association, Farm House Fraternity, Rocky Mountain Elk Foundation (Life Member), Pheasants Forever, Nature Conservancy, Cache Valley Wildlife Federation (and on Executive Board).

Current Teaching Assignment

NFS 5560 Chemistry of Food Systems
NFS 6450 Meat Science
NFS 1000 World of Food and Nutrition

I also assist with the NFS Food Science Club College Bowl team, and coordinate (with Dr. Carpenter) the Utah Future Farmers of America (FFA) state contest in Food Science and Technology.

Invited Speaker (1997-2001)

Cornforth, D. P. 1997. *Pigment concentration and pH effects on degree of doneness of beef patties*. FMC Corporation. Atlanta, Ga, January, 1997.

Cornforth, D. P. 1997. *Nitrogen dioxide causes surface pinking on meats cooked in gas ovens*. Meat Industry Meat Conf. Chicago, IL, Oct 28-29.

Cornforth, D. P., Jiang, C. and Mumford, B. 1998. *Effect of storage time, storage and cooking temperature, and holding time after cooking on pigment levels in beef patties*. Symposium, New Developments in Meat Processing, Amer. Meat Sci. Assn. National Meeting, Storrs, CT., June 28-July 2.

Cornforth, D. P. 2000. Keynote speaker. *Pinking in Cooked Poultry. Current situation and general theories*. Given at the Symposium on Pinking in Cooked Poultry, Center for Excellence in Poultry Science, University of Arkansas, Fayetteville, Oct 12-13, 1999. Co-sponsored by Tyson Foods, NewlyWed Foods, and KFC, Inc.

Cornforth, D. P. 2000. *Pinking in Cooked Poultry*. Michigan State University, E. Lansing, MI, Nov. 7.

Cornforth, D. P. 2000. *Pinking in Cooked Poultry*. Michigan Turkey Processors, Zeeland, MI, Nov. 8.

Cornforth, D. P. 2001. *Meat Color*. Dinner speaker for the joint meeting of the Southern Minnesota section of IFT and the Meat Processing Short Course sponsored by the Agricultural Utilization Research Institute, Southwest State University, Marshall, MN, April 25.

National Offices and Committees (1997-2001)

USDA Advisory Committee for the Safe Preparation of Ground Beef - 1997-8.

AMSA (American Meat Science Association)

Graduate Student Poster Competition committee, Reciprocal Meat Conference, 1998-2002.

Executive Board 2000-2002.

IFT Executive Committee, Muscle Foods Division, Institute of Food Technologists, 1998-99.

Chair-Elect, Muscle Foods Division, 2000-2001.

Chair, Muscle Foods Division, 2001-2002.

Executive Council, Bonneville Section, IFT (Councilor representing Bonneville Section at National Meetings), 1998-99.

Journal of Muscle Foods – Editorial Board, 1998-2002

University Committee Assignments

Committee for Laboratory Safety. 1997-2001.

Faculty Senate, 2000 – 2003.

Faculty Senate Executive Committee 2000 – 2001.

Grants (1997-2001)

Cornforth, D. P. 1997-98. Factors affecting hamburger degree of doneness. McDonald's Corp. Oak Brook, IL. \$8,480.

Cornforth, D. P. 1998. Evaluation of various dairy fractions as inhibitors of lipid oxidation in precooked meats. Western Dairy Foods Center, Logan, UT, \$9,560.

Cornforth, D. P. 1999. Verification of nutrient label claims on lowfat milk, lean hamburger, and breakfast cereals. Dr. Alan Luke (Alumni Donor), \$1,500.

Cornforth, D. P. and Carpenter, C. E. 1999-2000. Evaluation of carbon monoxide treatment in modified atmosphere or vacuum packaging to increase color stability of fresh beef. National Cattlemen's Beef Association, \$14,500.

Carpenter, C.E. and Cornforth, D. P. 1999-2000. Effect of consumer bias for beef color and packaging on eating satisfaction. National Cattlemen's Beef Association, \$15,000.

Cornforth, D. P. 1999-2002. Evaluation of antioxidant properties of milk powders in cooked meats. Glanbia Foods, Twin Falls, ID. \$30,000.

Cornforth, D. P. 2000-2002. New antioxidants in cooked meats. Utah Agric. Exp. Stat. \$15,000.

Bailey, D. and Dickinson, D. (Cornforth, D.P., co-investigator). 2001-2003. Traceability: A market opportunity or threat to the US meat industry. USDA-CREES. \$160,000.

Cornforth, D. P. 2001-2002. Dried whey minerals as an antioxidant in processed meats. Dairy Management Institute (DMI). \$97,500.

Graduate Committees Chaired (1997-2001)

Heaton, K. M. 1998. Minimum levels of nitrite, nitric oxide, and carbon monoxide causing pinking in cooked beef and turkey rolls. M. S. thesis, Utah State University, Logan, Utah. (Now Extension Agent, Garfield & Kane Counties, Utah).

Moiseev, I. V. 1998. Prevention of pink discoloration and microbial safety of rolls and patties made from dark-cutting beef, PhD dissertation, Utah State University, Logan, Utah. (Now lab chemist, Borden Foods, Columbus, OH).

Racz, J. M. 1998. An inoculated pack study using *Clostridium sporogenes* PA 3679 to determine the shelf stability of a vacuum packaged meat/vegetable stick. M. S. thesis, Utah State University, Logan, Utah. (Now lab chemist, Fresenius Medical, Ogden, UT).

Jayasingh, P. 2001. Dried milk minerals as an antioxidant in processed meats. Ph.D. dissertation, Utah State University, Logan, Utah (work in progress).

Product Development and Extension

Stew Sticks are currently in retail production by Utah Jerky, Inc., Ogden, UT.

Cornforth, D. P., Bailey, D., and McEvoy, R. 1997. Developed a video of ostrich slaughter and processing, used by the new plant in Filmore, UT for employee training, and also used by USU extension agents.

Refereed Publications (1997-2001)

- Moiseev, I. V. and Cornforth, D. P. 1997. Sodium hydroxide and sodium tripolyphosphate effects on bind strength and sensory characteristics of restructured beef rolls. *Meat Sci.* 45:53-60.
- Quinton, R. D., Cornforth, D. P., Hendricks, D. G., Brennand, C. P. and Su, Y. K. 1997. Acceptability and composition of some acidified meat and vegetable stick products. *J. Food Sci.* 62:1250-1254.
- Cornforth, D. P., Rabovitser, J. K., Ahuja, S., Wagner, J. C., Hanson, R., Cummings, B. and Chudnovsky, Y. 1998. Carbon monoxide, nitric oxide, and nitrogen dioxide levels in gas ovens related to surface pinking of cooked beef and turkey. *J. Agric. Food Chem.* 46:255-61.
- Lee, B., Hendricks, D. G. and Cornforth, D. P. 1998. Antioxidant effects of carnosine and phytic acid in a model beef system. *J. Food Sci.* 63:394-398.
- Lee, B., Hendricks, D. G. and Cornforth, D. P. 1998. Effect of sodium phytate, sodium pyrophosphate, and sodium tripolyphosphate on physicochemical characteristics of restructured beef. *Meat Sci.* 50:273-283.
- Moiseev, I. V. and Cornforth, D. P. 1999. Treatments for prevention of persistent pinking in dark-cutting beef patties. *J. Food Sci.* 64:738-43.
- Lee, B., Hendricks, D. G. and Cornforth, D. P. 1999. A comparison of carnosine and ascorbic acid on color and lipid stability in a ground beef patty model system. *Meat Sci.* 51:245-253.
- Heaton, K. M., Cornforth, D. P., Moiseev, I. V., Egbert, W. R. and Carpenter, C. E. 2000. Minimum sodium nitrite levels for pinking of various cooked meats as related to use of direct or indirect-dried soy isolates in poultry rolls. *Meat Sci.* 55:321-329.
- Carpenter, C. E., Cornforth, D. P., Whittier, D. 2001. Consumer preferences for beef color and packaging did not affect eating satisfaction. *Meat Science* 57:359-363.
- Jayasingh, P. Cornforth, D. P., Carpenter, C. E., and Whittier, D. 2001. Evaluation of carbon monoxide treatment in modified atmosphere packaging or vacuum packaging to increase color stability of fresh beef. *Meat Science*, In press.

Book Chapters

- Cornforth, D. P. 2000. Miscellaneous Colorants - Cured Meat. Unit F6.2 in *Current Protocols in Food Analytical Chemistry*. S. J. Schwartz, Ed. John Wiley & Sons, Inc. New York, NY.
- Cornforth, D. P. 2001. Potential use of phytate as an antioxidant in cooked meats. Ch. 11 in *Food Phytates*. R. Reddy and S. K. Sathe, Eds. Technomic Publ., Inc., Rowayton, CT.

ATTACHMENT 8

ENVIRON

August 27, 2001

Eric Greenberg
Ungaretti & Harris
3500 Three First National Plaza
Chicago, IL 60602-4283

Re: Generally recognized as safe ("GRAS") determination for carbon monoxide from Pactiv ActiveTech food-contact packaging.

Dear Eric:

This letter reports my assessment, based on my review of the available information, that the potential consumer exposure to carbon monoxide from the Pactiv ActiveTech food-contact packaging is safe, and is also generally recognized as safe ("GRAS"), under the Federal Food, Drug, and Cosmetic Act ("FDCA" or "the Act") when used under conditions defined in the attached GRAS determination document. First, the safety of carbon monoxide in this use is demonstrated by comparison of the U.S. Environmental Protection Agency ("USEPA") national ambient air quality standard ("NAAQS") for carbon monoxide with the estimated daily intake ("EDI") of carbon monoxide under the intended conditions of use of the product. Exposure to a substance generally is considered safe for its intended use if the EDI is a fraction of the allowable daily intake ("ADI"). Second, the GRAS status of carbon monoxide in this use is affirmed by demonstrating that the safety of carbon monoxide from Pactiv ActiveTech food-contact packaging material in its intended use is generally recognized.

My GRAS determination was based on scientific procedures as outlined in the U.S. Food and Drug Administration ("FDA") regulations (at §170.30(b)). This section requires that the same quantity and quality of scientific evidence is available and is reviewed as is required to obtain approval of the substance as a food additive. Moreover, in addition to requiring scientific evidence of safety (as with a food additive), a GRAS determination also requires that this scientific evidence of safety be generally known and accepted by experts qualified in the appropriate scientific and technical fields. This common knowledge requirement of a GRAS determination includes two elements: (1) the data and information relied upon to establish the scientific basis for safety must be generally available; and (2) there must be a basis to conclude that there is a consensus among qualified experts about the safety of the substance for its intended use.

Based on the scientific literature, studies, conclusions, and restrictions presented in the GRAS determination document, I regard the proposed uses of carbon monoxide from Pactiv ActiveTech packaging material as generally recognized as safe because the use will result in an exposure that is well below an acceptable exposure level for carbon monoxide.

ENVIRON International Corporation

4350 North Fairfax Drive, Suite 300 • Arlington, VA 22203 USA

Tel: (410) 703-5150 • Fax: (410) 703-5151

No evidence exists in the available information on carbon monoxide that demonstrates, or suggests reasonable grounds to suspect, a hazard to the public health when carbon monoxide is used at levels that might reasonably be expected from its proposed use as an additive in the Pactiv ActiveTech food-contact packaging material as defined in the GRAS determination document.

It is my opinion that other qualified and competent scientists reviewing the same publicly available data would reach the same scientific conclusion. Therefore, carbon monoxide, as an additive in the Pactiv ActiveTech food-contact packaging material, is generally recognized as safe.



Vasilios H. Frankos, Ph.D.
Principal, Health Sciences
ENVIRON International Corporation
Arlington, Virginia

VASILIOS H. FRANKOS, Ph.D.

EDUCATION

- 1977 Ph.D., Pharmacology and Toxicology, University of Maryland Pharmacy School
- 1973 M.S., Biology, University of Maryland
- 1970 B.A., Biology, University of Maryland

EXPERIENCE

Dr. Frankos is a Principal at ENVIRON Corporation and has over 20 years of experience in the toxicological and pharmacological evaluation of data used to assess the risks posed by foods and food additives, drugs, medical devices, cosmetics, pesticides, and environmental and occupational exposures. He has also been involved in the development of exposure and risk assessment methodology. Since joining ENVIRON, Dr. Frankos has led, contributed to, or managed hundreds of projects in these areas.

Foods and Food Additives:

Dr. Frankos has worked on a wide variety of projects evaluating the safety of foods, direct and indirect food additives, and food contaminants. As part of these food-related safety evaluations, he has developed strategies for testing new direct and indirect additives, evaluated toxicity test data to support safety determinations, prepared Generally Recognized as Safe (GRAS) reviews, performed exposure and risk assessments, and developed regulatory strategies. Dr. Frankos has also presented safety evaluations to the FDA on behalf of clients. Some of his major projects in the area of foods and food additives include:

- Provided ongoing FDA-related scientific and technical support to the Coalition for Safe Ceramicware (CSC). Performed a safety assessment of ceramic pitchers with glazes containing lead using data collected on migration of lead into a food simulant and into real foods. This assessment was submitted to the FDA as part of the CSC's comments on the FDA's proposed rule changing the action level for lead from ceramic pitchers.
- Developed direct food additive petitions and GRAS self-affirmation documents for numerous food additives including, novel fibers sources, an anti-caking agent, enzymes, sugars, and a major new class of food additives.
- Conducted a GRAS self-affirmation review, including an evaluation of safety data, of the first bioengineered food approved by the FDA. Presented this GRAS review to the FDA on behalf of the client, a major biotechnology company.
- Developed a GRAS affirmation document for a cellulose product, manufactured by a novel bacterial fermentation process, with proposed food use as a suspending/thickening agent. Designed, placed, and monitored preclinical toxicity studies required for FDA approval.
- Estimated doses posing no significant risk for chemicals that could potentially leach from packaging into food. Assessed the potential human exposure to these chemicals from migration from packaging into food. Compared the potential ingested dose to the acceptable daily intake (ADI) for these chemicals.

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- Petitioned the FDA to sanction expanded use in foods of an approved food additive. Prepared a review and update of existing toxicological literature on the material and estimated the increase in exposure likely to result from proposed new uses.
- Evaluated the carcinogenic risk associated with exposure to acrylamide residues in food and methylene chloride in decaffeinated tea.
- Addressed issues relating to FDA's regulation of polychlorinated biphenyl (PCB) residues. Examined whether tolerances for PCBs in fish could be reinterpreted for less chlorinated PCBs that have lower or no carcinogenic potency. Determined necessary research to establish differences in potency between PCBs.
- Developed an innovative exposure and safety assessment for a novel single cell protein (mycoprotein) meat substitute that has been submitted to the FDA for approval.
- Conducted a simulated FDA review of a food additive petition on a new artificial sweetener submitted to the FDA by the client's competitor. Review included critical evaluation of product chemistry, efficacy, estimates of human exposure, animal and human toxicology data, pharmacokinetics and metabolism information, and the basis for determining the acceptable daily intake of the sweetener.
- Performed a detailed evaluation of toxicity and carcinogenicity studies sponsored by a major drug company and studies from the published literature for the company's non-nutritive sweetener and assessed the toxicological significance to humans. Assisted in submission of a food additive petition. Provided continued regulatory support during the FDA review process.

Human and Veterinary Drugs, Medical Devices, and Cosmetics:

Dr. Frankos has provided scientific and regulatory guidance to clients in the human and veterinary drug, medical device, and cosmetic industries. He has been involved in identifying and assessing the risks to humans associated with exposure to constituents of these products. He has assisted clients in these industries in interacting with the FDA and has assisted them in complying with all aspects of FDA regulations. Some of his major projects in the areas of drugs, medical devices, and cosmetics include:

- Reviewed two large epidemiological studies on the differences in adverse drug reaction rates between two types of radiographic contrast media. Prepared a safety review document on animal and human literature on contrast media.
- Performed an independent evaluation of a New Drug Application (NDA) submission to the FDA, with emphasis on review of efficacy studies.
- Assisted the medical device manufacturer in complying with FDA's post-approval requirements for its device including compliance with the Medical Device Reporting (MDR) rule, submission of updates to the PMA application, and ensuring that all labeling and marketing materials are in compliance with FDA regulations. Designed a post-marketing clinical trial for the device to comply with FDA recommendations.
- Evaluated the potential carcinogenic risks to humans of an over-the-counter (OTC) medication that is applied to the skin. Prepared a report on these findings that was submitted to the FDA.

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- Prepared and submitted to the FDA a New Drug Application (NDA) for a drug that holds promise for dramatically decreasing the high percentage of reocclusion that occurs in angioplasty patients.
- Assisted a major pharmaceutical manufacturer in assessing potential health risks associated with a specific ingredient of various over-the-counter (OTC) drugs.
- Critically evaluated both published and unpublished studies on a psychoactive drug and rendered an opinion to the client on potential health effects of the drug and whether a no-observed-effect level (NOEL) had been established.
- Provided guidelines for subchronic testing to evaluate the safety for human use of an allergen desensitizer that was produced by polymerizing the allergen through a glutaraldehyde treatment.
- Analyzed the potential risk to humans resulting from the use of Furazolidone as an animal drug. Determined an estimate of this risk and presented the estimate to the FDA at a public hearing.
- Assisted a major manufacturer of veterinary drug products in developing an approach to dealing with mouse liver tumors and their usefulness as evidence of carcinogenicity.
- Reviewed and evaluated a New Animal Drug Application for FDA submission. Advised on the necessity of future studies.
- Assembled an expert panel to address the safety of an antimicrobial agent, extracted from a plant source, for use in oral hygiene products (e.g. toothpaste and oral rinse). The evaluation included a review of the preclinical and clinical toxicologic database, analysis of exposure, and determination of margin of safety associated with the proposed oral uses.
- Critically evaluated the evidence cited by FDA as the basis for considering nitrofurantoin animal drugs to be carcinogenic under the meaning of the Delaney Clause of the Federal Food, Drug, and Cosmetic Act.
- Reviewed toxicity data to be submitted in support of an Investigational Device Exemption (IDE) for an implantable medical device. Recommended and monitored the performance of supplemental tests, performed an exposure assessment of substances leaching from the device into the systemic circulation, characterized the risk to health from such exposures, and assisted in the presentation of these findings to the FDA.
- Evaluated toxicity data on the materials used in a device intended to be implanted in the abdominal cavity. Examined the adequacy of the existing data on the device material, the safety of the material used, and the safety of the proposed replacement material. Recommended studies to improve the data for submission to the FDA.
- Conducted a quantitative risk assessment on numerous color additives used in dermally applied cosmetics including an evaluation of toxicity, an analysis of exposure, and a determination of quantitative risks.
- Performed a hypothetical risk assessment for two colors used in cosmetics, based on the assumption that, if tested, they would produce tumors in rats. Demonstrated that such an outcome would still allow continued safe use of these colors in cosmetics.

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Pesticides:

Dr. Frankos has assisted U.S. and foreign manufacturers in obtaining EPA and California registration of agricultural, forestry, and homeowner use pesticide products. Some of his major projects in the area of pesticides include:

Reviewed EPA's assessment of dietary oncogenic risk of two fungicides and advised the manufacturers of additional data needed to perform a quantitative risk assessment.

- Designed and supervised a field study to estimate exposure to a pesticide during "worst case" application. The study monitored the application of the chemical, measured exposure of the user during various phases of application and determined the effect of protective clothing on exposure.
- Reviewed the results of an aquatic organism field monitoring study and its supporting laboratory data for a major manufacturer of agricultural chemicals.
- Evaluated toxicity and prepared a risk assessment for residues of a fungicide in imported wines. Counseled client on process necessary to receive an EPA import tolerance for the fungicide. Advised client on additional data needed to support a tolerance.
- Designed and monitored toxicology studies for a German firm required for registration of a plant growth promotor and assisted in submitting data to the EPA.
- For a West German pesticide manufacturing company wishing to purchase the patent rights to a new pesticide developed in the U.S., provided counsel on the acceptability of the available data to EPA and OECD and the further data needed to obtain a registration in the U.S.

Environmental and Occupational Exposures:

Dr. Frankos has directed numerous exposure and risk assessments involving hundreds of chemicals that have been associated with industrial processes, toxic waste or municipal incinerators, and hazardous waste sites. These assessments have used computerized models and include all routes of exposure. Some of his major projects in the areas of environmental and occupational exposures include:

- Performed a safety assessment for the consumption of drinking water in contact with a piece of equipment that could potentially release lead, including an estimation of 1990 baseline blood lead levels for four subpopulations using the Integrated Uptake/Biokinetic Model and a determination of the maximum acceptable concentration of lead in drinking water that was potentially in contact with the equipment. Collected and reviewed information on factors that affect the leachability of lead into drinking water.
- Assisted manufacturers of the plasticizer di-(2-ethylhexyl) phthalate (DEHP) in developing a method for estimating exposure resulting from the chemical's presence in polyvinyl chloride (PVC) consumer products such as vinyl-covered furniture, vinyl wallpaper, flooring, and shower curtains.
- Critically reviewed acrylamide's carcinogenic activity in Fischer 344 rats and designed a new cancer bioassay in rats that will improve risk assessments for acrylamide.

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- Reviewed the scientific basis for establishing safety factors needed to protect workers from reproductive toxicity associated with glycol ether exposure.
- Reevaluated the data from a National Cancer Institute (NCI) bioassay on dibutyltin diacetate to resolve discrepant interpretations by NCI and FDA.
- Reviewed FDA and National Toxicology Program (NTP) evaluations of the bioassay on dimethylterephthalate and rendered an opinion on the adequacy of the NTP and FDA decisions.
- Supported the EPA Office of Solid Waste in developing measures of inherent toxicity which can be combined with exposure estimates to provide a relative ranking for scheduling hazardous solid waste.
- Provided scientific litigation support for a municipality in which its waste treatment plant and landfill were contaminated with PCBs.
- Participated in a number of projects examining the developmental, embryotoxic, and teratogenic effects of lead observed in animals, including the effects of lead on the reproductive systems of male and female rats. Recommended a no-observed-adverse-effect level (NOAEL) for lead.

Exposure and Risk Assessment Methodology Development:

Dr. Frankos has participated in the development of new exposure and risk assessment methodologies for federal and state regulatory agencies. He has also been integral in the development of exposure and risk modeling software. Some of his major projects in these areas include:

- Developed a background document for the EPA on reproductive toxicity risk assessment for use in drafting interagency risk assessment guidelines.
- Directed development of a scheme for the EPA that allows severity of toxic effects to be incorporated into safety evaluations of EPA regulated products.
- Summarized and compiled comments received by EPA on their proposed guidelines (1985) for developmental toxicity assessment.
- Assisted in evaluating EPA's procedures for estimating safe short-term exposure limits and in developing an alternative method that could be uniformly used by the entire agency.
- Assisted in the development of criteria for listing chemicals as developmental toxicants under California's Proposition 65.
- Directed the development of a computer software system, ERMA (Exposure and Risk Modeling Assistant) that enables ENVIRON to provide high quality, scientifically defensible evaluations of potential exposures and resultant risk.

Before joining ENVIRON Corporation, Dr. Frankos held the following positions:

- Associate Director, Life Sciences Division, Clement Associates. In that position he had the following experience:
 - Supervised a staff of eight scientists who assessed the risk posed by environmental contaminants, occupational carcinogens, pesticides, drugs, commercial product

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constituents, and food additives. Many of these projects involved evaluating human and animal toxicology data for use in conducting human risk assessments. This position required management of time constraints, budgetary limitations, and personnel allocations in a manner that provided the client with a scientifically defensible document.

- Directed preparation of reports for industry clients that included "Safety Assessment Strategies for Feminine Hygiene Products"; "The Impact of a Proposed Salicylate Warning on the Risk Associated with Diseases and Conditions in Children"; and "A Proposed Mechanism of Action for a Carcinogenic Hair Dye Ingredient."
- Directed and prepared a report to OSHA entitled "Formaldehyde Risk Assessment for Occupationally Exposed Workers" and assisted in developing guidelines for interspecies data extrapolation for use by OSHA in its revised cancer policy.
- Provided litigation toxicity support to private and industry clients and assisted private concerns in the development of testing protocols for the purposes of fulfilling regulatory requirements.
- Served as expert reproductive toxicity witness in the House of Representative's hearing on the "Relationships of Exposure to Toxic Chemicals and Reproductive Impairment."
- Staff Science Advisor, Office of Health Affairs, Commissioners Office, Food and Drug Administration.
 - Provided professional scientific expertise in pharmacology, toxicology, biochemistry, and biology requisite to the effective accomplishment of the Agency's scientific overview and leadership function. Served as technical expert, scientific advisor, and liaison of the Commissioner at the Bureau level on matters relevant to toxicologic and pharmacologic safety assessment of toxic substances to which humans and animals are exposed.
 - Responsibilities included performing risk assessments on compounds (such as PCB's, methapyraline, caffeine, etc.) that could be utilized by the Commissioner's office in choosing between various regulatory options. Routinely reviewed toxicity data on compounds as diverse as caffeine, methylsalicylate, xylitol, and sodium nitrite. Provided litigation support and testified as expert toxicology witness at the Administrative Law Judge hearing on the safety of cyclamate.
 - Participated in the National Toxicology Program (NTP). Duties included review of agency nominations for toxicity testing by NTP, research planning for the regulatory needs of FDA, preparation of the Annual Carcinogen Report, and participation in numerous NTP workgroups.
 - Interagency and international initiatives included chairing the Interagency Regulatory Liaison Group (IRLG) Workgroup on Reproductive Toxicity Risk Assessment. Planned, coordinated, and published a three-day symposium entitled, "The Effects of Foods and Drugs on the Development and Function of the Nervous System." Chaired and organized a national workshop on Reproductive Toxicity Risk Assessment.
 - Served as Project Director of a contract with the Environment Teratology Information Computer Division, Oak Ridge National Research Laboratory to develop computerized teratology literature data extraction, indexing, and collation. This data base system is being

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integrated into the ETIC master computer file and will be manipulated through the INQUIRE data base management system.

- Senior Toxicologist, Division of Toxicology, Center for Food Safety and Applied Nutrition, Food and Drug Administration.
 - Responsible for the toxicologic evaluation of compounds of special interest to the agency. Required expert review of data on carcinogenicity, promotion, reproduction, teratology, and pharmacokinetics. Presented scientific evaluations to congressional, departmental, and agency directors. Represented the agency on sensitive scientific issues. Areas of emphasis included evaluation of toxicity data on artificial sweeteners (saccharin, cyclamate, xylitol, etc.), development of reproductive toxicity risk assessment criteria, pharmacokinetics evaluation of compounds (methylene chloride, sodium nitrite, saccharin, etc.), and risk assessment.
 - Directed the development of a PC computer network to be used by the Review Toxicologist of the FDA Bureau of Foods. This system allows direct access to toxicology data bases at NLM, Oak Ridge, Dialog, and extensive FDA internal data bases and full manipulation, storage, and collation of personalized literature data bases.
- Toxicologist, Division of Toxicology, Bureau of Foods, Food and Drug Administration.
 - Responsible for toxicologic review of substances used as direct and indirect food additives. Duties required discussions with manufacturers, formulators, toxicologists, universities, and other agencies on experimental procedures for showing safety and estimating risk.
 - Additional duties and accomplishments included primary involvement in planning, testing, and implementing new proposed regulations that direct the priority assessment review of all food additives. Duties included designing and implementing modern toxicity testing protocols, protocol quality parameters, criteria for utilizing toxicity data, and computer compatible toxicology test summarization forms. Co-led a 12 person toxicology cyclic review team. This endeavor was awarded a commendable service citation.
 - Served as the toxicology member on the Program Advisory Board for the FDA's effort to modernize the storage and retrieval of safety information in the Bureau of Foods. Over two million dollars was expended for the computerization and microfiling of all Bureau petition files. This system is now fully operational within the FDA and is called SIREN.
- Research Assistant, Department of Pharmacology and Drug Abuse, Maryland Psychiatric Research Center.
 - Laboratory responsibilities included the daily analysis of the catecholamines and their metabolites in the urine of patients under different drug therapies. Used column chromatography to separate the monoamines and their metabolites with subsequent fluorometric determinations of the amounts. Techniques involved using radioisotopic methods to determine monoamine oxidase kinetics, substrate K_m , and oxygen requirements and preparing mitochondrial isolates to study by oxygenographic assay and radioisotopic methods.
 - Clinical responsibilities included the collection, collation, and statistical analysis of data obtained by researchers in various disciplines, such as clinical and physiological psychology, biochemistry, and pharmacology. Assisted in the design, implementation,

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and statistical analysis of a study concerning the use of naloxone and cyclazocine as narcotic antagonists in a population of paroled drug addicts. Implementation of the study involved administering, scoring, and statistically analyzing psychodiagnostic and intelligence tests.

HONORS

- 1980 FDA Commendable Service Award for Direct Food Additive Cyclic Review
- 1969-70 Baltimore University Club Scholarship
- 1966-67 American Hellenic Education Scholarship

PROFESSIONAL ACTIVITIES AND MEMBERSHIPS

Invited presentation entitled "Developmental and Reproductive Toxicity Testing of Implantable Medical Devices." Presentation for: 1996 Summer Short Course Series on Medical Device Biocompatibility: From Material Screening to Final Product Testing. Sponsored by: Case Western Reserve University. July 18-19, 1996, Baltimore, Maryland.

Invited presentation entitled "Risk Assessment: What is it? How can I use it?" Regional Meeting of the Association of Official Analytical Chemists (AOAC), October 27, 1994, College Park, Maryland.

Invited presentation entitled "Testing Requirements for Medical Devices" Annual Genetic Toxicology Workshop, May 3-5, 1993, Rockville, Maryland.

Society of Regulatory Toxicology and Pharmacology 1990 -

Food and Drug Law Institute 1990 -

Guest Lecturer, University of Vermont Law School "Risk Assessment at the Law" June 4-14, 1990, Burlington, Vermont.

Invited presentation entitled "Review of Safety and Toxicity of Sanguinaria and Sanguinarine" Symposium on Sanguinaria, April 25, 1990, Toronto, Ontario, Canada.

Invited presentation entitled "Health Risks and Safety Precautions" PCB Compliance, Cleanup and Disposal, March 27-28, 1990, Toronto, Ontario, Canada.

Environmental Law Institute 1989 -

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Invited panel member "Weight of Evidence Considerations in Identifying Reproductive and Developmental Toxicants" Risk Assessment Issues in Developmental and Reproductive Toxicology, Sept. 18-19, 1989, Perkeley, California.

Invited presentation entitled "Health Risk and Safety Precautions" Current Issues in PCB Compliance, May 24-25, 1989, Toronto, Canada.

Regulatory Affairs Professionals Society 1988 -

Invited presentation entitled "Risk Assessment for Effects Other than Cancer" 1986 Conference for Food Protection, Aug. 17-20, 1986, Ann Arbor, Michigan.

Invited work group member at the EPA sponsored "Consensus Workshop on the Evaluation of Maternal and Developmental Toxicity" May 12-14, 1986, Rockville, Maryland.

Co-author of an invited paper entitled "Acrylonitrile as a Carcinogen: Research Needs for Better Risk Assessment" presented at the International Conference on "Occupational and Environmental Significance of Industrial Carcinogens" October 7-9, 1985, Milan, Italy.

Invited presentation entitled "FDA Perspectives on the use of Teratology Data for Human Risk Assessment" Symposium on "Risk Assessment for Developmental Toxicity" Annual meeting of the Society of Toxicology, March 13, 1984, Atlanta, Georgia.

Guest Lecturer and Expert Consultant to the Environmental Teratology Information Computer Division, Oak Ridge National Research Laboratory. 1982.

Expert Witness before the U.S. House of Representatives Committee on Science and Technology hearing on the "Relationship of Exposure to Toxic Chemicals and Reproductive Impairment," 1982.

Guest speaker, FDA National Scientific Health Professionals Meeting; spoke on the National Toxicology Program. 1981.

Member, National Toxicology Program Chemical Evaluation Committee. 1979-1982.

Member, National Toxicology Program Annual Carcinogen Report Workgroup. 1979-1982.

Invited workshop panel member, Workshop on Biological and Statistical Implications of the ED Study on Related Data Bases, Mt. Sterling, Ohio; sponsored by the Society of Toxicology for the National Center for Toxicological Research. 1981.

Chairman - Interagency Regulatory Liaison Group - Reproductive Toxicity Risk Assessment Group. 1980-1981.

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Chairman and Organizer of the IRLG - Reproductive Toxicity Risk Assessment Workshop held at FDA, Rockville, MD. 1981.

Participated in preparation of the National Science Foundation Third Annual Science and Technology Report. 1980.

Participated in Preparation of the National Science Foundation Five-Year Science Outlook. 1980.

Participated in publication of the First Annual Report on Carcinogens (DHHS/NTP). 1980.

Organizer and lecturer for a State Department and DHHS/FDA sponsored course conducted for the National Organization for Drug Control and Research (Cairo, Egypt), entitled: "A Course in Chronic and Reproductive Toxicity Testing and Risk Assessment." 1980.

Guest lecturer at FDA Consumer Exchange Meeting: Use of Risk Assessment for Decision-Making. 1980.

Co-chairman and organizer of the Fifth FDA Science Symposium on Effects of Foods and Drugs on the Development and Function of the Nervous System: Methods for Predicting Toxicity. Delivered presentation entitled "Symposium Summary and Future Directions for Neurotoxicology Testing." 1979.

Expert witness FDA Administrative Law Judge hearing on the Safety of Cyclamate Used as an Artificial Sweetener. 1979.

PUBLICATIONS

Kruger, C.L., M.H. Whittaker, and V.H. Frankos. 1999. Genotoxicity Tests on D-Tagatose. *Regulatory Toxicology and Pharmacology*. 29(2):S36-S42.

Kruger, C.L., M.H. Whittaker, and V.H. Frankos. 1999. Developmental Toxicity of D-Tagatose. *Regulatory Toxicology and Pharmacology*. 29(2):S29-S35.

Turnbull, D., M.H. Whittaker, V.H. Frankos, and D. Jonker. 1999. 90-Day Oral Toxicity Study of D-Tagatose in Rats. *Regulatory Toxicology and Pharmacology*. 29(2):S1-S10.

Trunbull, D., M.H. Whitaker, V.H. Frankos, and D. Jonker. 1999. 13-Week Oral Toxicity Study with Stanol Esters in Rats. *Regulatory Toxicology and Pharmacology*. 29(1):216-226.

Whittaker, M.H., V.H. Frankos, D.H. Waalkens-Berendsen, A.P.M. Wolterbeek. 1999. 2-Generation Reproductive Toxicity Study of Plant Stanol Esters in Rats. *Regulatory Toxicology and Pharmacology*. 29(1):196-204.

Hester, T.R., N.F. Ford, P.J. Gale, J.L. Hammett, R. Raymond, D. Turnbull, V.H. Frankos, and M.B. Cohen. 1997. Measurement of 2,4-toluenediamine in urine and serum samples from women with méme or replicon breast implants. *Plastic and Reconstructive Surgery*. 100(5):1296-1298.

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- Spiegel, J.E., R. Rose, P. Karabell, V.H. Frankos, and D.F. Schmitt. 1994. Safety and Benefits of Fructooligosaccharides as Food Ingredients. *Food Technology*. 85-89.
- Redenbaugh, K., T. Berner, D. Emlay, V. Frankos, W. Hiatt, C. Houck, M. Kramer, L. Malyj, B. Martineau, N. Rachman, L. Rudenko, R. Sanders, R. Sheehy, and R. Wixtrom. 1993. Regulatory Issues for Commercialization of Tomatoes with an Antisense Polygalacturonase Gene. *In Vitro Cell. Dev. Biol.* 29:17-26.
- Frankos, V.H., D.F. Schmitt, L.C. Haws, A.J. McEvily, R. Iyengar, S.A. Miller, I.C. Munro, F.M. Clydesdale, A.L. Forbes, and R.M. Sauer. 1991. Generally Recognized as Safe (GRAS) Evaluation of 4-Hexylresorcinol for Use as a Processing Aid for Prevention of Melanosis in Shrimp. *Regulatory Toxicology and Pharmacology* 14:202-212.
- Wilcock, K.E., A.B. Santamaria, V.H. Frankos, H.W. Fischer, F. Laden, E.A. Platz, and B.A. Jackson. 1990. Perspectives on Adverse Reaction Rates Associated with the Use of High Osmolar Ionic and Low Osmolar Nonionic Contrast Media. *Journal of the American College of Toxicology* 9(6):563-607.
- Frankos, V.H., D.J. Brusick, E.M. Johnson, H.I. Maibach, I. Munro, R.A. Squire, and C.S. Weil. 1990. Safety of Sanguinaria Extract as Used in Commercial Toothpaste and Oral Rinse Products. *Journal of the Canadian Dental Association* 56(7 Suppl):41-47.
- Schmitt, D., V. Frankos, and D. Richardson. 1990. Toxicologic Evaluation of Sanguinaria Extract. Eleventh Annual Meeting of the American College of Toxicology. Program and Abstracts. Abstract.
- Schmitt, D., V. Frankos, J. Westland, and T. Zoetis. 1990. Toxicological Evaluation of Cellulon™ Fiber: Genotoxicity, Acute and Subchronic Toxicity. Eleventh Annual Meeting of the American College of Toxicology. Program and Abstracts. Abstract.
- Rudenko, L., J. Adgate, M. Aponte-Pons, T. Berner, V. Frankos, R. Gregory, C.K. Lintner, N. Rachman, W. Sherman, T. Winters, and R. Wixtrom. 1990. Application of Risk Assessment Methodology to Genetically Engineered Food Products: A Generic Approach. Society for Risk Assessment. Abstract.

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- Hanson, C.F., V.H. Frankos, and W.O. Thompson. 1989. Bioavailability of oxalic acid from spinach, sugar beet fibre and a solution of sodium oxalate consumed by female volunteers. *Fd. Chem. Toxic.* 27,3:181-184.
- Frankos, V., L.H. Dulak, M.A. Fiedman. 1989. Use of risk assessment in the statistical design of a carcinogenicity bioassay of acrylamide. *The Toxicologist* 9:179. Abstract.
- Strother, D.E., R.W. Mast, R.C. Kraska, and V. Frankos. 1988. Acrylonitrile as a carcinogen. Research needs for a better risk assessment. *Annals of the New York Academy of Sciences* 534:169-178.
- Hanson, C.F., V.H. Frankos, and W.O. Thompson. 1988. Low dietary availability of oxalic acid present in refined sugar beet pulp compared to spinach and sodium oxalate. *The Toxicologist* 8:88. Abstract.
- Strother, D.E., R.W. Mast, R.C. Krashka, and V. Frankos. 1988. Acrylonitrile as a Carcinogen: Research Needs for Better Risk Assessment, *Annals of the New York Academy of Sciences* 534:169-178.
- Schwetz, A. Bernard, R.W. Tyl et al. 1987. Consensus workshop on the evaluation of maternal and developmental toxicity work group III report: Low dose extrapolation and other considerations for risk assessment - models and applications. *Teratogenesis, Carcinogenesis, and Mutagenesis* 7:321-327.
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- Flamm, W.G. and V. Frankos. 1985. Formaldehyde: Laboratory Evidence. In "Interpretation of Negative Epidemiological Evidence for Carcinogenicity." IARC Scientific Publication 65.

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- Siegel, D.M., V.H. Frankos, and M.A. Schneiderman. 1983. Formaldehyde risk assessment for occupationally exposed workers. *Regulatory Tox. and Pharm.* 3, 355-371.
- Frankos, V.H. 1982. Relationship of exposure to toxic chemicals and reproductive impairment. Expert testimony for Committee on Science and Technology, U.S. House of Representatives July 27, 1982. United State Government Printing Office.
- Frankos, V.H. 1982. Qualitative comparison of chemical teratogenesis in humans and animal species (Abstract). Third Annual Meeting of the American College of Toxicology, Washington, D.C.
- Siegel, D.M., V.H. Frankos, and M.A. Schneiderman. 1982. Formaldehyde risk assessment for occupationally exposed workers (Abstract). Third Annual Meeting of the American College of Toxicology, Washington, D.C.
- Frankos, V.H. and J. Wassom. 1982. Computerized teratology literature data extraction, indexing, and collation (Abstract). *Teratology*. 25:2 80A.
- Frankos, V.H. Symposium Summary and Suggested Future Directions for Detection of Neurotoxicity. 1980. In *The Effects of Foods and Drugs on the Development and Function of the Nervous System: Methods for Predicting Toxicity*. Edited by Gryder, R.M. and Frankos, V.H. U.S. Department of Health and Human Services, Food and Drug Administration, Publication No. DHHS/FDA 80-1076.
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ATTACHMENT 9

CURRICULUM VITAE

Navn/Name: Sørheim, Oddvin
Privatadresse/Private address: Lyngveien 24A, 1430 Ås
E-mail: oddvin.sorheim@matforsk.no
Telefon/Telephone: +47-64970100
Arbidssted/Place of employment: MATFORSK – Norwegian Food Research Institute
Osloveien 1, 1430 Ås
Stilling/Present position: Sjefingeniør / Senior Research Technologist
Dr.agric. / Ph.D.

Utdanning/Education:

2000, Dr. agric./Ph.D., Norges landbrukshøgskole/Agricultural University of Norway

1982, Cand. agric./M.Sc., Næringsmiddelfag, Norges landbrukshøgskole/Agricultural University of Norway

Erfering/Experience:

- 01.05. 1985 til nå ved/ to present at MATFORSK, arbeidet med forskning og industrioppdrag på kjøtt i hovedsak innen gasspakking, farge, mørhet, salting og funksjonelle egenskaper/ mainly working in research and industry consulting on packaging, color, tenderness, salting and functional properties of meat.
- 01.08. 1989 til 01.06. 1990, forskningsopphold/visiting scientist, Kansas State University, Department of Animal Sciences and Industry, Manhattan, KS, USA
- 01.06. 1982 til 30.04. 1985, produksjonskonsulent/consultant, Slakterienes Salgssentral/Norwegian Meat Cooperative, Oslo

Vitenskapelige originalarbeider/Scientific publications:

- Sørheim, O., Idland, J., Halvorsen, E.C., Frøystein, T., Lea, P., Hildrum, K.I. 2001. Influence of beef carcass stretching and chilling rate on the tenderness of *M. longissimus dorsi*. Meat Science, Vol 57, pp 79-85.
- Kjos, N.P., Øverland, M., Bryhni, E.A., Sørheim, O. 2000. Food waste products in diets for growing-finishing pigs: effect on growth performance, carcass characteristics and meat quality. Acta Agric. Scand., Sect. A, Animal Sci., Vol 50, pp 193-204.
- Hunt, M.C., Sørheim, O., Slinde, E. 1999. Color and heat denaturation of myoglobin forms in ground beef. Journal of Food Science, Vol 64, pp 847-851.
- Sørheim, O., Nissen, H., Nesbakken, T. 1999. The storage life of beef and pork packaged in an atmosphere with low carbon monoxide and high carbon dioxide. Meat Science, Vol 52, pp 157-164.
- Sørheim, O., Aune, T., Nesbakken, T. 1997. Technological, hygienic and toxicological aspects of carbon monoxide used in modified-atmosphere packaging of meat. Trends in Food Science & Technology, Vol 8, pp 307-312.
- Sørheim, O., Erlandsen, T., Nissen, H., Lea, P., Høyem, T. 1997. Effects of modified atmosphere storage on colour and microbiological shelf life of normal and pale, soft and exudative pork. Meat Science, Vol 47, pp 147-155.
- Sørheim, O., Kropf, D.H., Hunt, M.C., Yarwoski, M.T., Warren, K.E. 1996. Effects of modified gas atmosphere packaging on pork loin colour, display life and drip loss. Meat Science, Vol 43, pp 203-212.
- Nissen, H., Sørheim, O., Dainty, R.H. 1996. Effects of vacuum, modified atmospheres and storage temperature on the microbial flora of packaged beef. Food Microbiology, Vol 13, pp 183-191.
- Sørheim, O., Grini, J.Aa., Nissen, H., Andersen, H.J., Lea, P. 1995. Pork loins stored in carbon dioxide - Colour and microbiological shelf life. Fleischwirtschaft, 75, 679-681.

Vitenskapelige publikasjoner og presentasjoner/Other publications and presentations:

- Sørheim, O. 2000. Effects of modified atmosphere packaging on colour and microbiological shelf life of red meats. (Dr.agric./Ph.D.-thesis, Agricultural University of Norway).
- Sørheim, O., Nissen, H. 2000. Current technology of meat MAP. *Int. Food Marketing & Technology*, Vol 14, No 4, pp 39-42.
- Sørheim, O., Nissen, H. 1996. Modified atmosphere packaging of red meats. *The European Food & Drink Review*, No 4, pp 77-80.

Bok/book

- Hildrum, K.I., Dainty, R.H., Egelanddal, B., Larssen, E.G., Mielnik, J., Sørheim, O. 1996. Meat for the Consumer. MATFORSK Ås, 593 pp, ISBN 82-90394-58-6.

Annet/other

- Risvik, E., Hildrum, K.I., Russwurm jr, H., Dainty, R.H., Egelanddal, B., Green, A., Larssen, E.G., Merok, K.J., Mielnik, J., Norang, R.E., Næs, H., Olsen, M.Ø., Roskifte, P., Slinde, E., Sørheim, O. 1996. The 42nd ICoMST: "Meat for the Consumer", Norway 1-6 September 1996 (organising committee).
- Sørheim, O. 2001 - . Editorial board, *Journal of Muscle Foods*.

Proceedings:

- Sørheim, O., Nissen, H., Aune, T., Nesbakken, T. 2001. Use of carbon monoxide for retail meat packaging. *Proc. International Animal and Agriculture Food Science Conference, Indianapolis, USA*, in press.
- Sørheim, O., Lea, P., Nissen, H., Nesbakken, T. 2001. Effects of a high CO₂ /low CO atmosphere on colour and yield of cooked ground beef patties. *Proc. 47th ICoMST, Krakow, Poland*, 4 p., in press.
- Sørheim, O., Idland, J., Frøystein, T., Lea, P., Hildrum, K.I. 2000. Combined effects of aitch bone suspension and chilling rate on the tenderness of beef muscles. *Proc. 46th ICoMST, 27 August - 1 September 2000, Buenos Aires, Argentina*, Vol 1, pp 402-403.
- Bryhni, E.A., Kjos, N.P., Øverland, M., Sørheim, O. 1999. Food waste products in diets for growing-finishing pigs. Effect on growth performance, carcass characteristics and meat quality. *Proc. 45th ICoMST, 1-6 August 1999, (Eds. Japan Society for Meat Science and Technology) Yokohama, Japan*, pp 82-83.
- Sørheim, O., Nissen, H., Nesbakken, T. 1998. Color stabilization of pork chops packaged with a low level of carbon monoxide. *Proc. 51st Annual Reciprocal Meat Conference: Current Advances in Meat and Poultry Color, 28 June - 1 July 1998, (American Meat Science Association) Storrs, Connecticut, USA*, Vol 51, pp 187-188.
- Sørheim, O., Nissen, H., Nesbakken, T. 1997. Shelf life and colour stability of beef loin steaks packaged in a modified atmosphere with carbon monoxide., *Proc. 43rd ICoMST, 27 July - 1 August 1997, (Ed. J. Bass) Auckland, New Zealand*, Vol 43, pp 694-695. ISBN 0-473-04549-4.
- Sørheim, O., Lea, P., Gilde, M., Nissen, H. 1996. Effects of packaging gases on the colour of beef. *Proc. 42nd ICoMST (Ed. K.I. Hildrum), MATFORSK, Lillehammer, Norway, 1-6 September 1996*, pp 110-111, ISBN 82-90394-58-6.
- Hunt, M.C., Sørheim, O., Slinde, E. 1995. Effects of myoglobin form on internal cooked color development in ground beef. *Proc. 41st ICoMST, American Meat Science Association, San Antonio, Texas, USA, 20-25 August 1995*, Vol II, pp 394-395.

1 of 5 DOCUMENTS



Analysis
As of: May 15, 2007

KSR INTERNATIONAL CO., PETITIONER v. TELEFLEX INC. ET AL.

No. 04-1350

SUPREME COURT OF THE UNITED STATES

127 S. Ct. 1727; 2007 U.S. LEXIS 4745; 75 U.S.L.W. 4289

November 28, 2006, Argued
April 30, 2007, Decided

NOTICE: [*1] The LEXIS pagination of this document is subject to change pending release of the final published version.

PRIOR HISTORY: ON WRIT OF CERTIORARI TO THE UNITED STATES COURT OF APPEALS FOR THE FEDERAL CIRCUIT. *Teleflex, Inc. v. KSR Int'l Co.*, 119 Fed. Appx. 282, 2005 U.S. App. LEXIS 176 (Fed. Cir., 2005)

DISPOSITION: Reversed and remanded.

CASE SUMMARY:

PROCEDURAL POSTURE: Respondent, licensees of a patent, alleged that petitioner, a competitor, infringed the licensees' patent for an accelerator pedal assembly for vehicles, but the competitor asserted that the patent claim in dispute was invalid as obvious under 35 U.S.C.S. § 103. Upon the grant of a writ of certiorari, the competitor appealed the judgment of the U.S. Court of Appeals for the Federal Circuit which reversed a summary judgment of patent invalidity.

OVERVIEW: To satisfy customer needs, the competitor modified its design for an adjustable pedal system for vehicles with cable-actuated throttles by adding a modular sensor to make the system compatible with vehicles using computer-controlled throttles. The licensees contended that the competitor infringed the patent claim of a position-adjustable pedal assembly with an electronic pedal position sensor attached a fixed pivot point. The U.S. Supreme Court unanimously held that the patent

claim was invalid as obvious since mounting an available sensor on a fixed pivot point of the competitor's pedal was a design step well within the grasp of a person of ordinary skill in the relevant art, and the benefit of doing so was obvious. The marketplace created a strong incentive to convert mechanical pedals to electronic pedals, and the prior art taught a number of methods for doing so. Further, the problem to be solved by the patent claim did not limit its application as prior art, the competitor's showing that it was obvious to try a combination of elements sufficiently supported the finding of obviousness, and the claim was the result of ordinary skill and common sense rather than innovation.

OUTCOME: The judgment reversing the summary judgment of invalidity was reversed, and the case was remanded for further proceedings.

SYLLABUS: To control a conventional automobile's speed, the driver depresses or releases the gas pedal, which interacts with the throttle via a cable or other mechanical link. Because the pedal's position in the footwell normally cannot be adjusted, a driver wishing to be closer or farther from it must either reposition himself in the seat or move the seat, both of which can be imperfect solutions for smaller drivers in cars with deep footwells. This prompted inventors to design and patent pedals that could be adjusted to change their locations. The Asano patent reveals a support structure whereby, when the pedal location is [*2] adjusted, one of the pedal's pivot points stays fixed. Asano is also designed so that the force necessary to depress the pedal is the same regardless of location adjustments. The Redding patent reveals

a different, sliding mechanism where both the pedal and the pivot point are adjusted.

In newer cars, computer-controlled throttles do not operate through force transferred from the pedal by a mechanical link, but open and close valves in response to electronic signals. For the computer to know what is happening with the pedal, an electronic sensor must translate the mechanical operation into digital data. Inventors had obtained a number of patents for such sensors. The so-called '936 patent taught that it was preferable to detect the pedal's position in the pedal mechanism, not in the engine, so the patent disclosed a pedal with an electronic sensor on a pivot point in the pedal assembly. The Smith patent taught that to prevent the wires connecting the sensor to the computer from chafing and wearing out, the sensor should be put on a fixed part of the pedal assembly rather than in or on the pedal's footpad. Inventors had also patented self-contained modular sensors, which can be [*3] taken off the shelf and attached to any mechanical pedal to allow it to function with a computer-controlled throttle. The '068 patent disclosed one such sensor. Chevrolet also manufactured trucks using modular sensors attached to the pedal support bracket, adjacent to the pedal and engaged with the pivot shaft about which the pedal rotates. Other patents disclose electronic sensors attached to adjustable pedal assemblies. For example, the Rixon patent locates the sensor in the pedal footpad, but is known for wire chafing.

After petitioner KSR developed an adjustable pedal system for cars with cable-actuated throttles and obtained its '976 patent for the design, General Motors Corporation (GMC) chose KSR to supply adjustable pedal systems for trucks using computer-controlled throttles. To make the '976 pedal compatible with the trucks, KSR added a modular sensor to its design. Respondents (Teleflex) hold the exclusive license for the Engelgau patent, claim 4 of which discloses a position-adjustable pedal assembly with an electronic pedal position sensor attached a fixed pivot point. Despite having denied a similar, broader claim, the U.S. Patent and Trademark Office (PTO) had allowed [*4] claim 4 because it included the limitation of a fixed pivot position, which distinguished the design from Redding's. Asano was neither included among the Engelgau patent's prior art references nor mentioned in the patent's prosecution, and the PTO did not have before it an adjustable pedal with a fixed pivot point. After learning of KSR's design for GMC, Teleflex sued for infringement, asserting that KSR's pedal system infringed the Engelgau patent's claim 4. KSR countered that claim 4 was invalid under § 103 of the Patent Act, which forbids issuance of a patent when "the differences between the subject matter sought to be patented and the

prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art."

Graham v. John Deere Co. of Kansas City, 383 U.S. 1, 17-18, 86 S. Ct. 684, 15 L. Ed. 2d 545, set out an objective analysis for applying § 103: "The scope and content of the prior art are . . . determined; differences between the prior art and the claims at issue are . . . ascertained; and the level of ordinary skill in the pertinent art resolved. Against this background the obviousness or nonobviousness [*5] of the subject matter is determined. Such secondary considerations as commercial success, long felt but unsolved needs, failure of others, etc., might be utilized to give light to the circumstances surrounding the origin of the subject matter sought to be patented." While the sequence of these questions might be reordered in any particular case, the factors define the controlling inquiry. However, seeking to resolve the obviousness question with more uniformity and consistency, the Federal Circuit has employed a "teaching, suggestion, or motivation" (TSM) test, under which a patent claim is only proved obvious if the prior art, the problem's nature, or the knowledge of a person having ordinary skill in the art reveals some motivation or suggestion to combine the prior art teachings.

The District Court granted KSR summary judgment. After reviewing pedal design history, the Engelgau patent's scope, and the relevant prior art, the court considered claim 4's validity, applying *Graham's* framework to determine whether under summary-judgment standards KSR had demonstrated that claim 4 was obvious. The court found "little difference" between the prior art's teachings and claim 4: [*6] Asano taught everything contained in the claim except using a sensor to detect the pedal's position and transmit it to a computer controlling the throttle. That additional aspect was revealed in, e.g., the '068 patent and Chevrolet's sensors. The court then held that KSR satisfied the TSM test, reasoning (1) the state of the industry would lead inevitably to combinations of electronic sensors and adjustable pedals, (2) Rixon provided the basis for these developments, and (3) Smith taught a solution to Rixon's chafing problems by positioning the sensor on the pedal's fixed structure, which could lead to the combination of a pedal like Asano with a pedal position sensor.

Reversing, the Federal Circuit ruled the District Court had not applied the TSM test strictly enough, having failed to make findings as to the specific understanding or principle within a skilled artisan's knowledge that would have motivated one with no knowledge of the invention to attach an electronic control to the Asano assembly's support bracket. The Court of Appeals held

that the District Court's recourse to the nature of the problem to be solved was insufficient because, unless the prior art references [*7] addressed the precise problem that the patentee was trying to solve, the problem would not motivate an inventor to look at those references. The appeals court found that the Asano pedal was designed to ensure that the force required to depress the pedal is the same no matter how the pedal is adjusted, whereas Engelgau sought to provide a simpler, smaller, cheaper adjustable electronic pedal. The Rixon pedal, said the court, suffered from chafing but was not designed to solve that problem and taught nothing helpful to Engelgau's purpose. Smith, in turn, did not relate to adjustable pedals and did not necessarily go to the issue of motivation to attach the electronic control on the pedal assembly's support bracket. So interpreted, the court held, the patents would not have led a person of ordinary skill to put a sensor on an Asano-like pedal. That it might have been obvious to try that combination was likewise irrelevant. Finally, the court held that genuine issues of material fact precluded summary judgment.

Held: The Federal Circuit addressed the obviousness question in a narrow, rigid manner that is inconsistent with § 103 and this Court's precedents. KSR provided convincing [*8] evidence that mounting an available sensor on a fixed pivot point of the Asano pedal was a design step well within the grasp of a person of ordinary skill in the relevant art and that the benefit of doing so would be obvious. Its arguments, and the record, demonstrate that the Engelgau patent's claim 4 is obvious. Pp. 11-24.

1. *Graham* provided an expansive and flexible approach to the obviousness question that is inconsistent with the way the Federal Circuit applied its TSM test here. Neither § 103's enactment nor *Graham's* analysis disturbed the Court's earlier instructions concerning the need for caution in granting a patent based on the combination of elements found in the prior art. See *Great Atlantic & Pacific Tea Co. v. Supermarket Equipment Corp.*, 340 U.S. 147, 152, 71 S. Ct. 127, 95 L. Ed. 162, 1951 Dec. Comm'r Pat. 572. Such a combination of familiar elements according to known methods is likely to be obvious when it does no more than yield predictable results. See, e.g., *United States v. Adams*, 383 U.S. 39, 50-52, 86 S. Ct. 708, 15 L. Ed. 2d 572, 174 Ct. Cl. 1293. When a work is available in one field, design incentives and other market forces can prompt variations of it, either in the same field or in another. If a person [*9] of ordinary skill in the art can implement a predictable variation, and would see the benefit of doing so, § 103 likely bars its patentability. Moreover, if a technique has been used to improve one device, and a person of ordinary skill in the art would recognize that it would improve similar de-

vices in the same way, using the technique is obvious unless its actual application is beyond that person's skill. A court must ask whether the improvement is more than the predictable use of prior-art elements according to their established functions. Following these principles may be difficult if the claimed subject matter involves more than the simple substitution of one known element for another or the mere application of a known technique to a piece of prior art ready for the improvement. To determine whether there was an apparent reason to combine the known elements in the way a patent claims, it will often be necessary to look to interrelated teachings of multiple patents; to the effects of demands known to the design community or present in the marketplace; and to the background knowledge possessed by a person having ordinary skill in the art. To facilitate review, this analysis should [*10] be made explicit. But it need not seek out precise teachings directed to the challenged claim's specific subject matter, for a court can consider the inferences and creative steps a person of ordinary skill in the art would employ. Pp. 11-14.

(b) The TSM test captures a helpful insight: A patent composed of several elements is not proved obvious merely by demonstrating that each element was, independently, known in the prior art. Although common sense directs caution as to a patent application claiming as innovation the combination of two known devices according to their established functions, it can be important to identify a reason that would have prompted a person of ordinary skill in the art to combine the elements as the new invention does. Inventions usually rely upon building blocks long since uncovered, and claimed discoveries almost necessarily will be combinations of what, in some sense, is already known. Helpful insights, however, need not become rigid and mandatory formulas. If it is so applied, the TSM test is incompatible with this Court's precedents. The diversity of inventive pursuits and of modern technology counsels against confining the obviousness analysis [*11] by a formalistic conception of the words teaching, suggestion, and motivation, or by overemphasizing the importance of published articles and the explicit content of issued patents. In many fields there may be little discussion of obvious techniques or combinations, and market demand, rather than scientific literature, may often drive design trends. Granting patent protection to advances that would occur in the ordinary course without real innovation retards progress and may, for patents combining previously known elements, deprive prior inventions of their value or utility. Since the TSM test was devised, the Federal Circuit doubtless has applied it in accord with these principles in many cases. There is no necessary inconsistency between the test and the *Graham* analysis. But a court

errs where, as here, it transforms general principle into a rigid rule limiting the obviousness inquiry. Pp. 14-15.

(c) The flaws in the Federal Circuit's analysis relate mostly to its narrow conception of the obviousness inquiry consequent in its application of the TSM test. The Circuit first erred in holding that courts and patent examiners should look only to the problem the patentee was trying [*12] to solve. Under the correct analysis, any need or problem known in the field and addressed by the patent can provide a reason for combining the elements in the manner claimed. Second, the appeals court erred in assuming that a person of ordinary skill in the art attempting to solve a problem will be led only to those prior art elements designed to solve the same problem. The court wrongly concluded that because Asano's primary purpose was solving the constant ratio problem, an inventor considering how to put a sensor on an adjustable pedal would have no reason to consider putting it on the Asano pedal. It is common sense that familiar items may have obvious uses beyond their primary purposes, and a person of ordinary skill often will be able to fit the teachings of multiple patents together like pieces of a puzzle. Regardless of Asano's primary purpose, it provided an obvious example of an adjustable pedal with a fixed pivot point, and the prior art was replete with patents indicating that such a point was an ideal mount for a sensor. Third, the court erred in concluding that a patent claim cannot be proved obvious merely by showing that the combination of elements was obvious to try. [*13] When there is a design need or market pressure to solve a problem and there are a finite number of identified, predictable solutions, a person of ordinary skill in the art has good reason to pursue the known options within his or her technical grasp. If this leads to the anticipated success, it is likely the product not of innovation but of ordinary skill and common sense. Finally, the court drew the wrong conclusion from the risk of courts and patent examiners falling prey to hindsight bias. Rigid preventative rules that deny recourse to common sense are neither necessary under, nor consistent with, this Court's case law. Pp. 15-18.

2. Application of the foregoing standards demonstrates that claim 4 is obvious. Pp. 18-23.

(a) The Court rejects Teleflex's argument that the Asano pivot mechanism's design prevents its combination with a sensor in the manner claim 4 describes. This argument was not raised before the District Court, and it is unclear whether it was raised before the Federal Circuit. Given the significance of the District Court's finding that combining Asano with a pivot-mounted pedal position sensor fell within claim 4's scope, it is apparent that Teleflex would [*14] have made clearer challenges if it intended

to preserve this claim. Its failure to clearly raise the argument, and the appeals court's silence on the issue, lead this Court to accept the District Court's conclusion. Pp. 18-20.

(b) The District Court correctly concluded that when Engelgau designed the claim 4 subject matter, it was obvious to a person of ordinary skill in the art to combine Asano with a pivot-mounted pedal position sensor. There then was a marketplace creating a strong incentive to convert mechanical pedals to electronic pedals, and the prior art taught a number of methods for doing so. The Federal Circuit considered the issue too narrowly by, in effect, asking whether a pedal designer writing on a blank slate would have chosen both Asano and a modular sensor similar to the ones used in the Chevrolet trucks and disclosed in the '068 patent. The proper question was whether a pedal designer of ordinary skill in the art, facing the wide range of needs created by developments in the field, would have seen an obvious benefit to upgrading Asano with a sensor. For such a designer starting with Asano, the question was where to attach the sensor. The '936 patent taught [*15] the utility of putting the sensor on the pedal device. Smith, in turn, explained not to put the sensor on the pedal footpad, but instead on the structure. And from Rixon's known wire-chafing problems, and Smith's teaching that the pedal assemblies must not precipitate any motion in the connecting wires, the designer would know to place the sensor on a non-moving part of the pedal structure. The most obvious such point is a pivot point. The designer, accordingly, would follow Smith in mounting the sensor there. Just as it was possible to begin with the objective to upgrade Asano to work with a computer-controlled throttle, so too was it possible to take an adjustable electronic pedal like Rixon and seek an improvement that would avoid the wire-chafing problem. Teleflex has not shown anything in the prior art that taught away from the use of Asano, nor any secondary factors to dislodge the determination that claim 4 is obvious. Pp. 20-23.

3. The Court disagrees with the Federal Circuit's holding that genuine issues of material fact precluded summary judgment. The ultimate judgment of obviousness is a legal determination. *Graham*, 383 U.S., at 17, 86 S. Ct. 684, 15 L. Ed. 2d 545. Where, as here, the [*16] prior art's content, the patent claim's scope, and the level of ordinary skill in the art are not in material dispute and the claim's obviousness is apparent, summary judgment is appropriate. P. 23.

119 Fed. Appx. 282, reversed and remanded.

JUDGES: KENNEDY, J., delivered the opinion for a unanimous Court.

OPINION BY: KENNEDY**OPINION:**

JUSTICE KENNEDY delivered the opinion of the Court.

Teleflex Incorporated and its subsidiary Technology Holding Company -- both referred to here as Teleflex -- sued KSR International Company for patent infringement. The patent at issue, United States Patent No. 6,237,565 B1, is entitled "Adjustable Pedal Assembly With Electronic Throttle Control." Supplemental App. 1. The patentee is Steven J. Engelgau, and the patent is referred to as "the Engelgau patent." Teleflex holds the exclusive license to the patent.

Claim 4 of the Engelgau patent describes a mechanism for combining an electronic sensor with an adjustable automobile pedal so the pedal's position can be transmitted to a computer that controls the throttle in the vehicle's engine. When Teleflex accused KSR of infringing the Engelgau patent by adding an electronic sensor to one of KSR's previously [*17] designed pedals, KSR countered that claim 4 was invalid under the Patent Act, 35 U.S.C. § 103, because its subject matter was obvious.

Section 103 forbids issuance of a patent when "the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains."

In *Graham v. John Deere Co. of Kansas City*, 383 U.S. 1, 86 S. Ct. 684, 15 L. Ed. 2d 545 (1966), the Court set out a framework for applying the statutory language of § 103, language itself based on the logic of the earlier decision in *Hotchkiss v. Greenwood*, 52 U.S. 248, 11 How. 248, 13 L. Ed. 683 (1851), and its progeny. See 383 U.S., at 15-17, 86 S. Ct. 684, 15 L. Ed. 2d 545. The analysis is objective:

"Under § 103, the scope and content of the prior art are to be determined; differences between the prior art and the claims at issue are to be ascertained; and the level of ordinary skill in the pertinent art resolved. Against this background the obviousness or nonobviousness of the subject matter is determined. Such secondary considerations [*18] as commercial success, long felt but unsolved needs, failure of others, etc., might be utilized to give light to the circumstances surrounding the origin of the subject matter sought to be

patented." *Id.*, at 17-18, 86 S. Ct. 684, 15 L. Ed. 2d 545.

While the sequence of these questions might be reordered in any particular case, the factors continue to define the inquiry that controls. If a court, or patent examiner, conducts this analysis and concludes the claimed subject matter was obvious, the claim is invalid under § 103.

Seeking to resolve the question of obviousness with more uniformity and consistency, the Court of Appeals for the Federal Circuit has employed an approach referred to by the parties as the "teaching, suggestion, or motivation" test (TSM test), under which a patent claim is only proved obvious if "some motivation or suggestion to combine the prior art teachings" can be found in the prior art, the nature of the problem, or the knowledge of a person having ordinary skill in the art. See, e.g., *Al-Site Corp. v. VSI Int'l, Inc.*, 174 F.3d 1308, 1323-1324 (CA Fed. 1999). KSR challenges that test, or at least its application in this case. See 119 Fed. Appx. 282, 286-290 (CA Fed. 2005). [*19] Because the Court of Appeals addressed the question of obviousness in a manner contrary to § 103 and our precedents, we granted certiorari, 547 U.S. , 126 S. Ct. 2965, 165 L. Ed. 2d 949 (2006). We now reverse.

I

A

In car engines without computer-controlled throttles, the accelerator pedal interacts with the throttle via cable or other mechanical link. The pedal arm acts as a lever rotating around a pivot point. In a cable-actuated throttle control the rotation caused by pushing down the pedal pulls a cable, which in turn pulls open valves in the carburetor or fuel injection unit. The wider the valves open, the more fuel and air are released, causing combustion to increase and the car to accelerate. When the driver takes his foot off the pedal, the opposite occurs as the cable is released and the valves slide closed.

In the 1990's it became more common to install computers in cars to control engine operation. Computer-controlled throttles open and close valves in response to electronic signals, not through force transferred from the pedal by a mechanical link. Constant, delicate adjustments of air and fuel mixture are possible. The computer's rapid processing of factors beyond the pedal's position improves [*20] fuel efficiency and engine performance.

For a computer-controlled throttle to respond to a driver's operation of the car, the computer must know what is happening with the pedal. A cable or mechanical

link does not suffice for this purpose; at some point, an electronic sensor is necessary to translate the mechanical operation into digital data the computer can understand.

Before discussing sensors further we turn to the mechanical design of the pedal itself. In the traditional design a pedal can be pushed down or released but cannot have its position in the footwell adjusted by sliding the pedal forward or back. As a result, a driver who wishes to be closer or farther from the pedal must either reposition himself in the driver's seat or move the seat in some way. In cars with deep footwells these are imperfect solutions for drivers of smaller stature. To solve the problem, inventors, beginning in the 1970's, designed pedals that could be adjusted to change their location in the footwell. Important for this case are two adjustable pedals disclosed in U.S. Patent Nos. 5,010,782 (filed July 28, 1989) (Asano) and 5,460,061 (filed Sept. 17, 1993) (Redding). The Asano patent reveals a [*21] support structure that houses the pedal so that even when the pedal location is adjusted relative to the driver, one of the pedal's pivot points stays fixed. The pedal is also designed so that the force necessary to push the pedal down is the same regardless of adjustments to its location. The Redding patent reveals a different, sliding mechanism where both the pedal and the pivot point are adjusted.

We return to sensors. Well before Engelgau applied for his challenged patent, some inventors had obtained patents involving electronic pedal sensors for computer-controlled throttles. These inventions, such as the device disclosed in U.S. Patent No. 5,241,936 (filed Sept. 9, 1991) ('936), taught that it was preferable to detect the pedal's position in the pedal assembly, not in the engine. The '936 patent disclosed a pedal with an electronic sensor on a pivot point in the pedal assembly. U.S. Patent No. 5,063,811 (filed July 9, 1990) (Smith) taught that to prevent the wires connecting the sensor to the computer from chafing and wearing out, and to avoid grime and damage from the driver's foot, the sensor should be put on a fixed part of the pedal assembly rather than in or on the pedal's [*22] footpad.

In addition to patents for pedals with integrated sensors inventors obtained patents for self-contained modular sensors. A modular sensor is designed independently of a given pedal so that it can be taken off the shelf and attached to mechanical pedals of various sorts, enabling the pedals to be used in automobiles with computer-controlled throttles. One such sensor was disclosed in U.S. Patent No. 5,385,068 (filed Dec. 18, 1992) ('068). In 1994, Chevrolet manufactured a line of trucks using modular sensors "attached to the pedal support bracket, adjacent to the pedal and engaged with the pivot shaft about which the pedal rotates in operation." 298 F. Supp. 2d 581, 589 (ED Mich. 2003).

The prior art contained patents involving the placement of sensors on adjustable pedals as well. For example, U.S. Patent No. 5,819,593 (filed Aug. 17, 1995) (Rixon) discloses an adjustable pedal assembly with an electronic sensor for detecting the pedal's position. In the Rixon pedal the sensor is located in the pedal footpad. The Rixon pedal was known to suffer from wire chafing when the pedal was depressed and released.

This short account of pedal and sensor technology leads [*23] to the instant case.

B

KSR, a Canadian company, manufactures and supplies auto parts, including pedal systems. Ford Motor Company hired KSR in 1998 to supply an adjustable pedal system for various lines of automobiles with cable-actuated throttle controls. KSR developed an adjustable mechanical pedal for Ford and obtained U.S. Patent No. 6,151,976 (filed July 16, 1999) ('976) for the design. In 2000, KSR was chosen by General Motors Corporation (GMC or GM) to supply adjustable pedal systems for Chevrolet and GMC light trucks that used engines with computer-controlled throttles. To make the '976 pedal compatible with the trucks, KSR merely took that design and added a modular sensor.

Teleflex is a rival to KSR in the design and manufacture of adjustable pedals. As noted, it is the exclusive licensee of the Engelgau patent. Engelgau filed the patent application on August 22, 2000 as a continuation of a previous application for U.S. Patent No. 6,109,241, which was filed on January 26, 1999. He has sworn he invented the patent's subject matter on February 14, 1998. The Engelgau patent discloses an adjustable electronic pedal described in the specification as a "simplified vehicle control [*24] pedal assembly that is less expensive, and which uses fewer parts and is easier to package within the vehicle." Engelgau, col. 2, lines 2-5, Supplemental App. 6. Claim 4 of the patent, at issue here, describes:

"A vehicle control pedal apparatus comprising:

a support adapted to be mounted to a vehicle structure;

an adjustable pedal assembly having a pedal arm moveable in fore and aft directions with respect to said support;

a pivot for pivotally supporting said adjustable pedal assembly with respect to said support and defining a pivot axis; and

an electronic control attached to said support for controlling a vehicle system;

said apparatus characterized by said electronic control being responsive to said pivot for providing a signal that corresponds to pedal arm position as said pedal arm pivots about said pivot axis between rest and applied positions wherein the position of said pivot remains constant while said pedal arm moves in fore and aft directions with respect to said pivot." *Id.*, col. 6, lines 17-36, Supplemental App. 8 (diagram numbers omitted).

We agree with the District Court that the claim discloses "a position-adjustable pedal [*25] assembly with an electronic pedal position sensor attached to the support member of the pedal assembly. Attaching the sensor to the support member allows the sensor to remain in a fixed position while the driver adjusts the pedal." 298 F. Supp. 2d, at 586-587.

Before issuing the Engelgau patent the U.S. Patent and Trademark Office (PTO) rejected one of the patent claims that was similar to, but broader than, the present claim 4. The claim did not include the requirement that the sensor be placed on a fixed pivot point. The PTO concluded the claim was an obvious combination of the prior art disclosed in Redding and Smith, explaining:

"Since the prior art references are from the field of endeavor, the purpose disclosed . . . would have been recognized in the pertinent art of Redding. Therefore it would have been obvious . . . to provide the device of Redding with the . . . means attached to a support member as taught by Smith." *Id.*, at 595.

In other words Redding provided an example of an adjustable pedal and Smith explained how to mount a sensor on a pedal's support structure, and the rejected patent claim merely put these two teachings together. [*26]

Although the broader claim was rejected, claim 4 was later allowed because it included the limitation of a fixed pivot point, which distinguished the design from Redding's. *Ibid.* Engelgau had not included Asano among the prior art references, and Asano was not mentioned in the patent's prosecution. Thus, the PTO did not have before it an adjustable pedal with a fixed pivot point. The patent issued on May 29, 2001 and was assigned to Teleflex.

Upon learning of KSR's design for GM, Teleflex sent a warning letter informing KSR that its proposal would violate the Engelgau patent. "Teleflex believes that any supplier of a product that combines an adjustable pedal with an electronic throttle control necessarily employs technology covered by one or more" of Teleflex's patents. *Id.*, at 585. KSR refused to enter a royalty arrangement with Teleflex; so Teleflex sued for infringement, asserting KSR's pedal infringed the Engelgau patent and two other patents. *Ibid.* Teleflex later abandoned its claims regarding the other patents and dedicated the patents to the public. The remaining contention was that KSR's pedal system for GM infringed claim 4 of the Engelgau patent. [*27] Teleflex has not argued that the other three claims of the patent are infringed by KSR's pedal, nor has Teleflex argued that the mechanical adjustable pedal designed by KSR for Ford infringed any of its patents.

C

The District Court granted summary judgment in KSR's favor. After reviewing the pertinent history of pedal design, the scope of the Engelgau patent, and the relevant prior art, the court considered the validity of the contested claim. By direction of 35 U.S.C. § 282, an issued patent is presumed valid. The District Court applied *Graham's* framework to determine whether under summary-judgment standards KSR had overcome the presumption and demonstrated that claim 4 was obvious in light of the prior art in existence when the claimed subject matter was invented. See § 102(a).

The District Court determined, in light of the expert testimony and the parties' stipulations, that the level of ordinary skill in pedal design was "an undergraduate degree in mechanical engineering (or an equivalent amount of industry experience) [and] familiarity with pedal control systems for vehicles." 298 F. Supp. 2d, at 590. The court then set forth the [*28] relevant prior art, including the patents and pedal designs described above.

Following *Graham's* direction, the court compared the teachings of the prior art to the claims of Engelgau. It found "little difference." 298 F. Supp. 2d, at 590. Asano taught everything contained in claim 4 except the use of a sensor to detect the pedal's position and transmit it to the computer controlling the throttle. That additional aspect was revealed in sources such as the '068 patent and the sensors used by Chevrolet.

Under the controlling cases from the Court of Appeals for the Federal Circuit, however, the District Court was not permitted to stop there. The court was required also to apply the TSM test. The District Court held KSR had satisfied the test. It reasoned (1) the state of the industry would lead inevitably to combinations of electronic sensors and adjustable pedals, (2) Rixon provided

the basis for these developments, and (3) Smith taught a solution to the wire chafing problems in Rixon, namely locating the sensor on the fixed structure of the pedal. This could lead to the combination of Asano, or a pedal like it, with a pedal position sensor.

The conclusion that the [*29] Engelgau design was obvious was supported, in the District Court's view, by the PTO's rejection of the broader version of claim 4. Had Engelgau included Asano in his patent application, it reasoned, the PTO would have found claim 4 to be an obvious combination of Asano and Smith, as it had found the broader version an obvious combination of Redding and Smith. As a final matter, the District Court held that the secondary factor of Teleflex's commercial success with pedals based on Engelgau's design did not alter its conclusion. The District Court granted summary judgment for KSR.

With principal reliance on the TSM test, the Court of Appeals reversed. It ruled the District Court had not been strict enough in applying the test, having failed to make "findings as to the specific understanding or principle within the knowledge of a skilled artisan that would have motivated one with no knowledge of [the] invention' . . . to attach an electronic control to the support bracket of the Asano assembly." 119 Fed. Appx., at 288 (brackets in original) (quoting *In re Kotzab*, 217 F.3d 1365, 1371 (CA Fed. 2000)). The Court of Appeals held that the District Court was [*30] incorrect that the nature of the problem to be solved satisfied this requirement because unless the "prior art references addressed the precise problem that the patentee was trying to solve," the problem would not motivate an inventor to look at those references. 119 Fed. Appx., at 288.

Here, the Court of Appeals found, the Asano pedal was designed to solve the "constant ratio problem" -- that is, to ensure that the force required to depress the pedal is the same no matter how the pedal is adjusted -- whereas Engelgau sought to provide a simpler, smaller, cheaper adjustable electronic pedal. *Ibid.* As for Rixon, the court explained, that pedal suffered from the problem of wire chafing but was not designed to solve it. In the court's view Rixon did not teach anything helpful to Engelgau's purpose. Smith, in turn, did not relate to adjustable pedals and did not "necessarily go to the issue of motivation to attach the electronic control on the support bracket of the pedal assembly." *Ibid.* When the patents were interpreted in this way, the Court of Appeals held, they would not have led a person of ordinary skill to put a sensor on the sort of pedal described in Asano. [*31]

That it might have been obvious to try the combination of Asano and a sensor was likewise irrelevant, in the court's view, because " 'obvious to try' has long been

held not to constitute obviousness." *Id.*, at 289 (quoting *In re Deuel*, 51 F.3d 1552, 1559 (CA Fed. 1995)).

The Court of Appeals also faulted the District Court's consideration of the PTO's rejection of the broader version of claim 4. The District Court's role, the Court of Appeals explained, was not to speculate regarding what the PTO might have done had the Engelgau patent mentioned Asano. Rather, the court held, the District Court was obliged first to presume that the issued patent was valid and then to render its own independent judgment of obviousness based on a review of the prior art. The fact that the PTO had rejected the broader version of claim 4, the Court of Appeals said, had no place in that analysis.

The Court of Appeals further held that genuine issues of material fact precluded summary judgment. Teleflex had proffered statements from one expert that claim 4 "was a simple, elegant, and novel combination of features," 119 Fed. Appx., at 290, compared to Rixon, [*32] and from another expert that claim 4 was nonobvious because, unlike in Rixon, the sensor was mounted on the support bracket rather than the pedal itself. This evidence, the court concluded, sufficed to require a trial.

II

A

We begin by rejecting the rigid approach of the Court of Appeals. Throughout this Court's engagement with the question of obviousness, our cases have set forth an expansive and flexible approach inconsistent with the way the Court of Appeals applied its TSM test here. To be sure, *Graham* recognized the need for "uniformity and definiteness." 383 U.S., at 18, 86 S. Ct. 684, 15 L. Ed. 2d 545. Yet the principles laid down in *Graham* reaffirmed the "functional approach" of *Hotchkiss*, 52 U.S. 248, 11 How. 248, 13 L. Ed. 683. See 383 U.S., at 12, 86 S. Ct. 684, 15 L. Ed. 2d 545. To this end, *Graham* set forth a broad inquiry and invited courts, where appropriate, to look at any secondary considerations that would prove instructive. *Id.*, at 17, 86 S. Ct. 684, 15 L. Ed. 2d 545.

Neither the enactment of § 103 nor the analysis in *Graham* disturbed this Court's earlier instructions concerning the need for caution in granting a patent based on the combination of elements found in the prior art. For over a half century, [*33] the Court has held that a "patent for a combination which only unites old elements with no change in their respective functions . . . obviously withdraws what is already known into the field of its monopoly and diminishes the resources available to skillful men." *Great Atlantic & Pacific Tea Co. v. Supermarket Equipment Corp.*, 340 U.S. 147, 152, 71 S. Ct. 127, 95 L. Ed. 162, 1951 Dec. Comm'r Pat. 572 (1950). This is a principal reason for declining to allow patents

for what is obvious. The combination of familiar elements according to known methods is likely to be obvious when it does no more than yield predictable results. Three cases decided after *Graham* illustrate the application of this doctrine.

In *United States v. Adams*, 383 U.S. 39, 40, 86 S. Ct. 708, 15 L. Ed. 2d 572, 174 Ct. Cl. 1293 (1966), a companion case to *Graham*, the Court considered the obviousness of a "wet battery" that varied from prior designs in two ways: It contained water, rather than the acids conventionally employed in storage batteries; and its electrodes were magnesium and cuprous chloride, rather than zinc and silver chloride. The Court recognized that when a patent claims a structure already known in the prior art that is altered by the mere substitution of one [*34] element for another known in the field, the combination must do more than yield a predictable result. 383 U.S., at 50-51, 86 S. Ct. 708, 15 L. Ed. 2d 572, 174 Ct. Cl. 1293. It nevertheless rejected the Government's claim that Adams's battery was obvious. The Court relied upon the corollary principle that when the prior art teaches away from combining certain known elements, discovery of a successful means of combining them is more likely to be nonobvious. *Id.*, at 51-52, 86 S. Ct. 708, 15 L. Ed. 2d 572, 174 Ct. Cl. 1293. When Adams designed his battery, the prior art warned that risks were involved in using the types of electrodes he employed. The fact that the elements worked together in an unexpected and fruitful manner supported the conclusion that Adams's design was not obvious to those skilled in the art.

In *Anderson's-Black Rock, Inc. v. Pavement Salvage Co.*, 396 U.S. 57, 90 S. Ct. 305, 24 L. Ed. 2d 258 (1969), the Court elaborated on this approach. The subject matter of the patent before the Court was a device combining two pre-existing elements: a radiant-heat burner and a paving machine. The device, the Court concluded, did not create some new synergy: The radiant-heat burner functioned just as a burner was expected to function; and the paving machine did [*35] the same. The two in combination did no more than they would in separate, sequential operation. *Id.*, at 60-62, 90 S. Ct. 305, 24 L. Ed. 2d 258. In those circumstances, "while the combination of old elements performed a useful function, it added nothing to the nature and quality of the radiant-heat burner already patented," and the patent failed under § 103. *Id.*, at 62, 90 S. Ct. 305, 24 L. Ed. 2d 258 (footnote omitted).

Finally, in *Sakraida v. AG Pro, Inc.*, 425 U.S. 273, 96 S. Ct. 1532, 47 L. Ed. 2d 784 (1976), the Court derived from the precedents the conclusion that when a patent "simply arranges old elements with each performing the same function it had been known to perform" and yields no more than one would expect from such an ar-

rangement, the combination is obvious. *Id.*, at 282, 96 S. Ct. 1532, 47 L. Ed. 2d 784.

The principles underlying these cases are instructive when the question is whether a patent claiming the combination of elements of prior art is obvious. When a work is available in one field of endeavor, design incentives and other market forces can prompt variations of it, either in the same field or a different one. If a person of ordinary skill can implement a predictable variation, § 103 likely bars its patentability. For [*36] the same reason, if a technique has been used to improve one device, and a person of ordinary skill in the art would recognize that it would improve similar devices in the same way, using the technique is obvious unless its actual application is beyond his or her skill. *Sakraida* and *Anderson's-Black Rock* are illustrative -- a court must ask whether the improvement is more than the predictable use of prior art elements according to their established functions.

Following these principles may be more difficult in other cases than it is here because the claimed subject matter may involve more than the simple substitution of one known element for another or the mere application of a known technique to a piece of prior art ready for the improvement. Often, it will be necessary for a court to look to interrelated teachings of multiple patents; the effects of demands known to the design community or present in the marketplace; and the background knowledge possessed by a person having ordinary skill in the art, all in order to determine whether there was an apparent reason to combine the known elements in the fashion claimed by the patent at issue. To facilitate review, this analysis [*37] should be made explicit. See *In re Kahn*, 441 F.3d 977, 988 (CA Fed. 2006) ("Rejections on obviousness grounds cannot be sustained by mere conclusory statements; instead, there must be some articulated reasoning with some rational underpinning to support the legal conclusion of obviousness"). As our precedents make clear, however, the analysis need not seek out precise teachings directed to the specific subject matter of the challenged claim, for a court can take account of the inferences and creative steps that a person of ordinary skill in the art would employ.

B

When it first established the requirement of demonstrating a teaching, suggestion, or motivation to combine known elements in order to show that the combination is obvious, the Court of Customs and Patent Appeals captured a helpful insight. See *Application of Bergel*, 292 F.2d 955, 956-957, 48 C.C.P.A. 1102, 1961 Dec. Com'm'r Pat. 504 (1961). As is clear from cases such as *Adams*, a patent composed of several elements is not proved obvious merely by demonstrating that each of its elements was, independently, known in the prior art. Al-

though common sense directs one to look with care at a patent application that claims as innovation [*38] the combination of two known devices according to their established functions, it can be important to identify a reason that would have prompted a person of ordinary skill in the relevant field to combine the elements in the way the claimed new invention does. This is so because inventions in most, if not all, instances rely upon building blocks long since uncovered, and claimed discoveries almost of necessity will be combinations of what, in some sense, is already known.

Helpful insights, however, need not become rigid and mandatory formulas; and when it is so applied, the TSM test is incompatible with our precedents. The obviousness analysis cannot be confined by a formalistic conception of the words teaching, suggestion, and motivation, or by overemphasis on the importance of published articles and the explicit content of issued patents. The diversity of inventive pursuits and of modern technology counsels against limiting the analysis in this way. In many fields it may be that there is little discussion of obvious techniques or combinations, and it often may be the case that market demand, rather than scientific literature, will drive design trends. Granting patent protection [*39] to advances that would occur in the ordinary course without real innovation retards progress and may, in the case of patents combining previously known elements, deprive prior inventions of their value or utility.

In the years since the Court of Customs and Patent Appeals set forth the essence of the TSM test, the Court of Appeals no doubt has applied the test in accord with these principles in many cases. There is no necessary inconsistency between the idea underlying the TSM test and the *Graham* analysis. But when a court transforms the general principle into a rigid rule that limits the obviousness inquiry, as the Court of Appeals did here, it errs.

C

The flaws in the analysis of the Court of Appeals relate for the most part to the court's narrow conception of the obviousness inquiry reflected in its application of the TSM test. In determining whether the subject matter of a patent claim is obvious, neither the particular motivation nor the avowed purpose of the patentee controls. What matters is the objective reach of the claim. If the claim extends to what is obvious, it is invalid under § 103. One of the ways in which a patent's subject matter can be proved obvious is [*40] by noting that there existed at the time of invention a known problem for which there was an obvious solution encompassed by the patent's claims.

The first error of the Court of Appeals in this case was to foreclose this reasoning by holding that courts and patent examiners should look only to the problem the

patentee was trying to solve. 119 Fed. Appx., at 288. The Court of Appeals failed to recognize that the problem motivating the patentee may be only one of many addressed by the patent's subject matter. The question is not whether the combination was obvious to the patentee but whether the combination was obvious to a person with ordinary skill in the art. Under the correct analysis, any need or problem known in the field of endeavor at the time of invention and addressed by the patent can provide a reason for combining the elements in the manner claimed.

The second error of the Court of Appeals lay in its assumption that a person of ordinary skill attempting to solve a problem will be led only to those elements of prior art designed to solve the same problem. *Ibid.* The primary purpose of Asano was solving the constant ratio problem; so, the court concluded, [*41] an inventor considering how to put a sensor on an adjustable pedal would have no reason to consider putting it on the Asano pedal. *Ibid.* Common sense teaches, however, that familiar items may have obvious uses beyond their primary purposes, and in many cases a person of ordinary skill will be able to fit the teachings of multiple patents together like pieces of a puzzle. Regardless of Asano's primary purpose, the design provided an obvious example of an adjustable pedal with a fixed pivot point; and the prior art was replete with patents indicating that a fixed pivot point was an ideal mount for a sensor. The idea that a designer hoping to make an adjustable electronic pedal would ignore Asano because Asano was designed to solve the constant ratio problem makes little sense. A person of ordinary skill is also a person of ordinary creativity, not an automaton.

The same constricted analysis led the Court of Appeals to conclude, in error, that a patent claim cannot be proved obvious merely by showing that the combination of elements was "obvious to try." *Id.*, at 289 (internal quotation marks omitted). When there is a design need or market pressure to solve a problem [*42] and there are a finite number of identified, predictable solutions, a person of ordinary skill has good reason to pursue the known options within his or her technical grasp. If this leads to the anticipated success, it is likely the product not of innovation but of ordinary skill and common sense. In that instance the fact that a combination was obvious to try might show that it was obvious under § 103.

The Court of Appeals, finally, drew the wrong conclusion from the risk of courts and patent examiners falling prey to hindsight bias. A factfinder should be aware, of course, of the distortion caused by hindsight bias and must be cautious of arguments reliant upon *ex post* reasoning. See *Graham*, 383 U.S., at 36, 86 S. Ct. 684, 15 L. Ed. 2d 545 (warning against a "temptation to read into

the prior art the teachings of the invention in issue" and instructing courts to "guard against slipping into the use of hindsight" (quoting *Monroe Auto Equipment Co. v. Heckethorn Mfg. & Supply Co.*, 332 F.2d 406, 412 (CA6 1964))). Rigid preventative rules that deny factfinders recourse to common sense, however, are neither necessary under our case law nor consistent with it.

We note the [*43] Court of Appeals has since elaborated a broader conception of the TSM test than was applied in the instant matter. See, e.g., *DyStar Textilfarben GmbH & Co. Deutschland KG v. C. H. Patrick Co.*, 464 F.3d 1356, 1367 (2006) ("Our suggestion test is in actuality quite flexible and not only permits, but requires, consideration of common knowledge and common sense"); *Alza Corp. v. Mylan Labs., Inc.*, 464 F.3d 1286, 1291 (2006) ("There is flexibility in our obviousness jurisprudence because a motivation may be found implicitly in the prior art. We do not have a rigid test that requires an actual teaching to combine . . ."). Those decisions, of course, are not now before us and do not correct the errors of law made by the Court of Appeals in this case. The extent to which they may describe an analysis more consistent with our earlier precedents and our decision here is a matter for the Court of Appeals to consider in its future cases. What we hold is that the fundamental misunderstandings identified above led the Court of Appeals in this case to apply a test inconsistent with our patent law decisions.

III

When we apply the standards we have [*44] explained to the instant facts, claim 4 must be found obvious. We agree with and adopt the District Court's recitation of the relevant prior art and its determination of the level of ordinary skill in the field. As did the District Court, we see little difference between the teachings of Asano and Smith and the adjustable electronic pedal disclosed in claim 4 of the Engelgau patent. A person having ordinary skill in the art could have combined Asano with a pedal position sensor in a fashion encompassed by claim 4, and would have seen the benefits of doing so.

A

Teleflex argues in passing that the Asano pedal cannot be combined with a sensor in the manner described by claim 4 because of the design of Asano's pivot mechanisms. See Brief for Respondents 48-49, and n. 17. Therefore, Teleflex reasons, even if adding a sensor to Asano was obvious, that does not establish that claim 4 encompasses obvious subject matter. This argument was not, however, raised before the District Court. There Teleflex was content to assert only that the problem motivating the invention claimed by the Engelgau patent would not lead to the solution of combining of Asano with a sensor. See Teleflex's Response [*45] to KSR's

Motion for Summary Judgment of Invalidity in No. 02-74586 (ED Mich.), pp. 18-20, App. 144a-146a. It is also unclear whether the current argument was raised before the Court of Appeals, where Teleflex advanced the non-specific, conclusory contention that combining Asano with a sensor would not satisfy the limitations of claim 4. See Brief for Plaintiffs-Appellants in No. 04-1152 (CA Fed.), pp. 42-44. Teleflex's own expert declarations, moreover, do not support the point Teleflex now raises. See Declaration of Clark J. Radcliffe, Ph.D., Supplemental App. 204-207; Declaration of Timothy L. Andresen, *id.*, at 208-210. The only statement in either declaration that might bear on the argument is found in the Radcliffe declaration:

Asano . . . and Rixon . . . are complex mechanical linkage-based devices that are expensive to produce and assemble and difficult to package. It is exactly these difficulties with prior art designs that [Engelgau] resolves. The use of an adjustable pedal with a single pivot reflecting pedal position combined with an electronic control mounted between the support and the adjustment assembly at that pivot was a simple, elegant, and novel combination [*46] of features in the Engelgau '565 patent." *Id.*, at 206, P16.

Read in the context of the declaration as a whole this is best interpreted to mean that Asano could not be used to solve "the problem addressed by Engelgau '565[:] to provide a less expensive, more quickly assembled, and smaller package adjustable pedal assembly with electronic control." *Id.*, at 205, P10.

The District Court found that combining Asano with a pivot-mounted pedal position sensor fell within the scope of claim 4. 298 F. Supp. 2d, at 592-593. Given the significance of that finding to the District Court's judgment, it is apparent that Teleflex would have made clearer challenges to it if it intended to preserve this claim. In light of Teleflex's failure to raise the argument in a clear fashion, and the silence of the Court of Appeals on the issue, we take the District Court's conclusion on the point to be correct.

B

The District Court was correct to conclude that, as of the time Engelgau designed the subject matter in claim 4, it was obvious to a person of ordinary skill to combine Asano with a pivot-mounted pedal position sensor. There then existed a marketplace that created a strong [*47] incentive to convert mechanical pedals to electronic ped-

als, and the prior art taught a number of methods for achieving this advance. The Court of Appeals considered the issue too narrowly by, in effect, asking whether a pedal designer writing on a blank slate would have chosen both Asano and a modular sensor similar to the ones used in the Chevrolet truckline and disclosed in the '068 patent. The District Court employed this narrow inquiry as well, though it reached the correct result nevertheless. The proper question to have asked was whether a pedal designer of ordinary skill, facing the wide range of needs created by developments in the field of endeavor, would have seen a benefit to upgrading Asano with a sensor.

In automotive design, as in many other fields, the interaction of multiple components means that changing one component often requires the others to be modified as well. Technological developments made it clear that engines using computer-controlled throttles would become standard. As a result, designers might have decided to design new pedals from scratch; but they also would have had reason to make pre-existing pedals work with the new engines. Indeed, upgrading its [*48] own pre-existing model led KSR to design the pedal now accused of infringing the Engelgau patent.

For a designer starting with Asano, the question was where to attach the sensor. The consequent legal question, then, is whether a pedal designer of ordinary skill starting with Asano would have found it obvious to put the sensor on a fixed pivot point. The prior art discussed above leads us to the conclusion that attaching the sensor where both KSR and Engelgau put it would have been obvious to a person of ordinary skill.

The '936 patent taught the utility of putting the sensor on the pedal device, not in the engine. Smith, in turn, explained to put the sensor not on the pedal's footpad but instead on its support structure. And from the known wire-chafing problems of Rixon, and Smith's teaching that "the pedal assemblies must not precipitate any motion in the connecting wires," Smith, col. 1, lines 35-37, Supplemental App. 274, the designer would know to place the sensor on a nonmoving part of the pedal structure. The most obvious nonmoving point on the structure from which a sensor can easily detect the pedal's position is a pivot point. The designer, accordingly, would follow Smith [*49] in mounting the sensor on a pivot, thereby designing an adjustable electronic pedal covered by claim 4.

Just as it was possible to begin with the objective to upgrade Asano to work with a computer-controlled throttle, so too was it possible to take an adjustable electronic pedal like Rixon and seek an improvement that would avoid the wire-chafing problem. Following similar steps to those just explained, a designer would learn from Smith to avoid sensor movement and would come,

thereby, to Asano because Asano disclosed an adjustable pedal with a fixed pivot.

Teleflex indirectly argues that the prior art taught away from attaching a sensor to Asano because Asano in its view is bulky, complex, and expensive. The only evidence Teleflex marshals in support of this argument, however, is the Radcliffe declaration, which merely indicates that Asano would not have solved Engelgau's goal of making a small, simple, and inexpensive pedal. What the declaration does not indicate is that Asano was somehow so flawed that there was no reason to upgrade it, or pedals like it, to be compatible with modern engines. Indeed, Teleflex's own declarations refute this conclusion. Dr. Radcliffe states that [*50] Rixon suffered from the same bulk and complexity as did Asano. See *id.*, at 206. Teleflex's other expert, however, explained that Rixon was itself designed by adding a sensor to a pre-existing mechanical pedal. See *id.*, at 209. If Rixon's base pedal was not too flawed to upgrade, then Dr. Radcliffe's declaration does not show Asano was either. Teleflex may have made a plausible argument that Asano is inefficient as compared to Engelgau's preferred embodiment, but to judge Asano against Engelgau would be to engage in the very hindsight bias Teleflex rightly urges must be avoided. Accordingly, Teleflex has not shown anything in the prior art that taught away from the use of Asano.

Like the District Court, finally, we conclude Teleflex has shown no secondary factors to dislodge the determination that claim 4 is obvious. Proper application of *Graham* and our other precedents to these facts therefore leads to the conclusion that claim 4 encompassed obvious subject matter. As a result, the claim fails to meet the requirement of § 103.

We need not reach the question whether the failure to disclose Asano during the prosecution of Engelgau voids the presumption of validity given [*51] to issued patents, for claim 4 is obvious despite the presumption. We nevertheless think it appropriate to note that the rationale underlying the presumption -- that the PTO, in its expertise, has approved the claim -- seems much diminished here.

IV

A separate ground the Court of Appeals gave for reversing the order for summary judgment was the existence of a dispute over an issue of material fact. We disagree with the Court of Appeals on this point as well. To the extent the court understood the *Graham* approach to exclude the possibility of summary judgment when an expert provides a conclusory affidavit addressing the question of obviousness, it misunderstood the role expert testimony plays in the analysis. In considering summary judgment on that question the district court can and

should take into account expert testimony, which may resolve or keep open certain questions of fact. That is not the end of the issue, however. The ultimate judgment of obviousness is a legal determination. *Graham*, 383 U.S., at 17, 86 S. Ct. 684, 15 L. Ed. 2d 545. Where, as here, the content of the prior art, the scope of the patent claim, and the level of ordinary skill in the art are not in material dispute, and [*52] the obviousness of the claim is apparent in light of these factors, summary judgment is appropriate. Nothing in the declarations proffered by Teleflex prevented the District Court from reaching the careful conclusions underlying its order for summary judgment in this case.

* * *

We build and create by bringing to the tangible and palpable reality around us new works based on instinct, simple logic, ordinary inferences, extraordinary ideas, and sometimes even genius. These advances, once part of our shared knowledge, define a new threshold from which innovation starts once more. And as progress beginning from higher levels of achievement is expected in the normal course, the results of ordinary innovation are not the subject of exclusive rights under the patent laws. Were it otherwise patents might stifle, rather than promote, the progress of useful arts. See U.S. Const., Art. I,

§ 8, cl. 8. These premises led to the bar on patents claiming obvious subject matter established in *Hotchkiss* and codified in § 103. Application of the bar must not be confined within a test or formulation too constrained to serve its purpose.

KSR provided convincing evidence that mounting a modular [*53] sensor on a fixed pivot point of the Asano pedal was a design step well within the grasp of a person of ordinary skill in the relevant art. Its arguments, and the record, demonstrate that claim 4 of the Engelgau patent is obvious. In rejecting the District Court's rulings, the Court of Appeals analyzed the issue in a narrow, rigid manner inconsistent with § 103 and our precedents. The judgment of the Court of Appeals is reversed, and the case remanded for further proceedings consistent with this opinion.

It is so ordered.

REFERENCES: Go To Full Text Opinion

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United States Court of Appeals for the Federal Circuit

06-1402

LEAPFROG ENTERPRISES, INC.,

Plaintiff-Appellant,

v.

FISHER-PRICE, INC. and MATTEL, INC.,

Defendants-Appellees.

Ron E. Shulman, Wilson Sonsini Goodrich & Rosati, of Palo Alto, California, argued for plaintiff-appellant. With him on the brief were Terry Kearney and Michael A. Berta.

James Galbraith, Kenyon & Kenyon LLP, of New York, New York, argued for defendants-appellees. With him on the brief were Richard L. DeLucia and John Flock; and John R. Hutchins, of Washington, DC. Of counsel was Jeffrey M. Butler, of New York, New York.

Appealed from: United States District Court for the District of Delaware

Judge Gregory M. Sleet

United States Court of Appeals for the Federal Circuit

06-1402

LEAPFROG ENTERPRISES, INC.,

Plaintiff-Appellant,

v.

FISHER-PRICE, INC. and MATTEL, INC.,

Defendants-Appellees.

DECIDED: May 9, 2007

Before MAYER, LOURIE, and DYK, Circuit Judges.

LOURIE, Circuit Judge.

Leapfrog Enterprises, Inc. ("Leapfrog") appeals from the order of the United States District Court for the District of Delaware entering judgment of noninfringement and invalidity of claim 25 of Leapfrog's U.S. Patent 5,813,861 ("the '861 patent") in favor of Fisher-Price, Inc. and Mattel, Inc. (collectively "Fisher-Price"). We affirm.

BACKGROUND

Leapfrog filed suit in October 2003, alleging that Fisher-Price's PowerTouch product infringed claim 25 of the '861 patent. Leapfrog amended the complaint to add

Mattel, Inc. as a codefendant in September 2004. The '861 patent relates to a learning device to help young children read phonetically. Claim 25 reads as follows:

An interactive learning device, comprising:

- a housing including a plurality of switches;
- a sound production device in communication with the switches and including a processor and a memory;
- at least one depiction of a sequence of letters, each letter being associable with a switch; and
- a reader configured to communicate the identity of the depiction to the processor,

wherein selection of a depicted letter activates an associated switch to communicate with the processor, causing the sound production device to generate a signal corresponding to a sound associated with the selected letter, the sound being determined by a position of the letter in the sequence of letters.

'861 patent, col.10 ll.23-36.

In an April 7, 2005 Order, the trial court construed a number of terms from claim 25 of the patent. The court construed the phrase "selection of a depicted letter" to mean "choosing a particular depicted letter from the depicted sequence of letters by contacting or coming into proximity to that particular depicted letter." Leapfrog Enters., Inc. v. Fisher-Price, Inc., No. 03-927 (D. Del. Apr. 7, 2005).

The accused PowerTouch device consists of a hinged plastic housing containing electronics and a speaker that opens to lie flat. When so opened, a user places a book made for use with the device in a rectangular recess in the housing. The books contain large, colorful pictures that also show words associated with the objects shown in those pictures. The user may select one of multiple modes of operation. In phonics mode, when the user touches one of the words on the page, the device pronounces the word, then pronounces each phoneme of the word in sequence, and finally pronounces the

entire word again. The device relies on a grid of "crosspoints" located in the area underneath where the books are placed to detect the location on the page being touched by the user. The processor in the device may be programmed to associate a particular response with each crosspoint. Some of the words on the pages of the books are large enough that each letter of the word corresponds to a separate crosspoint. However, the phonics mode operates in the same manner for those words, with pronunciation of the word, the phonemes, and the word again, regardless which letter the user touches because each letter has been associated with the same response in the device's programming.

The case proceeded to trial, but the jury deadlocked on May 27, 2005. The parties stipulated that the case would be submitted to the trial court for decision, based on the record and the rulings made by the court at the time the case was submitted to the jury.

The trial court issued its decision on March 30, 2006, finding claim 25 of the '861 patent not infringed and invalid as obvious. The court found that the accused PowerTouch device could not practice the "selection of a depicted letter" because it only allowed selection of words rather than letters. The court thus found that the PowerTouch did not infringe claim 25. The court also concluded that claim 25 was invalid as obvious in view of the combination of U.S. Patent 3,748,748 to Bevan, the Texas Instruments Super Speak & Read ("SSR") device, and the knowledge of one of ordinary skill in the art as represented by the testimony of Fisher-Price's technical expert, Ronald Milner.

Leapfrog timely appealed. We have jurisdiction pursuant to 28 U.S.C. § 1295(a)(1).

DISCUSSION

A. Noninfringement

The district court's determination of infringement is a question of fact that we review for clear error. Abraxis Bioscience, Inc. v. Mayne Pharm. (USA) Inc., 467 F.3d 1370, 1375 (Fed. Cir. 2006). "Under the clear error standard, the court's findings will not be overturned in the absence of a definite and firm conviction that a mistake has been made." Impax Labs., Inc. v. Aventis Pharm. Inc., 468 F.3d 1366, 1375 (Fed. Cir. 2006) (quotation omitted).

On appeal, Leapfrog does not challenge the district court's construction of the phrase "selection of a depicted letter," but argues that the court clearly erred in applying that construction to the facts of the case. More specifically, Leapfrog argues that the PowerTouch does allow "choosing a particular depicted letter" because in at least some cases each letter of a word corresponds to a separate crosspoint. Thus, the fact that the response of the device is the same, no matter which letter the user touches, is irrelevant because the user may still choose particular letters.

Fisher-Price also does not challenge the district court's claim construction, and Fisher-Price responds that the district court correctly determined that selection by choosing a particular letter is only meaningful if making one letter choice results in an outcome different from making a different letter choice. Fisher-Price argues that the district court correctly found that only the word can be selected if the choice of letter, within a particular word, is irrelevant to the response of the device.

We find no clear error in the district court's application of the claim to the essentially undisputed facts of this case. The court's conclusion that the Fisher-Price PowerTouch only allows selection of a word rather than "a depicted letter" comports with its construction of "selection" to mean "choosing." The ordinary meaning of choice requires that the alternatives from which the choice is made will result in different possible outcomes. With the PowerTouch device, the same outcome results no matter which letter in the word the user touches. This understanding is also consistent with the way that selection of a depicted letter is described in the patent.

Every time the child depresses a letter key, the book will recite the phoneme of the letter associated with that letter, in the context that the letter is used in the word or phrase depicted on the card, here "ball." Thus, for the example where the subject is "ball" as shown if the child depresses the correct letter key of "b" the processor will sound the phoneme "b" as "b" is pronounced in "ball."

'861 patent, col.6 ll.17-23. Most importantly, this understanding of selection is also most consistent with the language of claim 25 itself. The PowerTouch device does not generate a signal corresponding to a sound associated with the selected letter, as the claim requires. A signal corresponding to a word is not the same as a signal corresponding to a letter. If the claim were meant to encompass a device that always enunciates all the letters of a word no matter which letter was selected, the claim language requiring that "the sound be[] determined by a position of the letter in the sequence of letters" would be superfluous because no such determination would be necessary.

Leapfrog comes well short of supporting a definite and firm conviction that a mistake has been made, and we therefore affirm the district court's entry of judgment of noninfringement in favor of Fisher-Price.

B. Obviousness

"Obviousness is a question of law, reviewed de novo, based upon underlying factual questions which are reviewed for clear error following a bench trial." Alza Corp. v. Mylan Labs., Inc., 464 F.3d 1286, 1289 (Fed. Cir. 2006) (citing Ruiz v. A.B. Chance Co., 357 F.3d 1270, 1275 (Fed. Cir. 2004)).

Leapfrog argues that the district court engaged in improper hindsight in reaching its conclusion of obviousness by concluding that all of the limitations of the claim are found in the prior art. Leapfrog also argues that the court's finding that the Bevan device has the same functionality as claim 25 was clearly erroneous because the components of Bevan's device are mechanical, and thus different in structure and interrelation from the electronic components described in claim 25, and therefore cannot provide the same functionality. Leapfrog argues that there was inadequate evidence in the record to support a motivation to combine Bevan, the Texas Instruments SSR, and a reader to arrive at the invention of claim 25. Finally, Leapfrog argues that the district court did not properly consider the strong evidence of secondary considerations of nonobviousness.

In response, Fisher-Price argues that claim 25 is nothing more than the Bevan device, a toy that teaches reading based on the association of letters with their phonemic sounds, updated with modern electronics that were common by the time of the alleged invention. Fisher-Price also responds that particularized and specific motivations to combine need not be found in the prior art references themselves in the context of an improvement that arises from a desire to generally improve a known device (e.g., to make the product smaller, lighter, or less expensive) using newer

technology. Finally, Fisher-Price argues that the district court did give proper consideration to secondary considerations of nonobviousness, but simply concluded that those considerations were not sufficient to overcome the determination of obviousness based on primary considerations.

We agree with Fisher-Price that the district court correctly concluded that the subject matter of claim 25 of the '861 patent would have been obvious in view of the combination of Bevan, the SSR, and the knowledge of one of ordinary skill in the art. An obviousness determination is not the result of a rigid formula disassociated from the consideration of the facts of a case. Indeed, the common sense of those skilled in the art demonstrates why some combinations would have been obvious where others would not. See KSR Int'l Co. v. Teleflex Inc., 550 U.S. ___, 2007 WL 1237837, at *12 (2007) ("The combination of familiar elements according to known methods is likely to be obvious when it does no more than yield predictable results."). Thus, we bear in mind that the goal of the claim 25 device was to allow a child to press a switch associated with a single letter in a word and hear the sound of the letter as it is used in that word. In this way, the child would both associate the sound of the letter with the letter itself and be able to sound out the word one letter at a time to learn to read phonetically. Accommodating a prior art mechanical device that accomplishes that goal to modern electronics would have been reasonably obvious to one of ordinary skill in designing children's learning devices. Applying modern electronics to older mechanical devices has been commonplace in recent years.

The Bevan patent was one of the pieces of prior art relied upon by the district court, and it describes an electro-mechanical learning toy. In the preferred embodiment

of the Bevan device, a housing contains a phonograph record as a voice storage means, a speaker for playing sounds from the voice storage means, and an actuated electric motor to turn the record. Uniquely shaped puzzle pieces fit into correspondingly shaped openings in the top of the housing. Depressing the puzzle pieces in the openings causes the motor to turn the record and brings phonographic needles into contact with the portions of the record where the sounds associated with the puzzle pieces are stored so that they can be played through the speaker. In one embodiment, each puzzle piece is imprinted with one letter from a word, and pressing each puzzle piece produces the sound of a single letter in that word. Thus, although it relies on an electric motor and mechanical structures rather than a processor and related electronics, Bevan teaches an apparatus that achieves the goals described above of associating letters with their sounds and encouraging children to sound out words phonetically through a similar type of interaction. We therefore see no clear error in the district court's finding that the Bevan device has the same method of operation, viewed as a whole, as claim 25 of Leapfrog's '861 patent.

A second piece of prior art relied upon by the district court was the Texas Instruments SSR. The SSR is a more modern type of prior art learning toy, constructed with electronic components, that has a slightly different mode of operation than Bevan. The SSR has a hinged plastic housing that opens to lie flat. Books for use with the toy fit into a recess in the housing. The housing contains switches that can detect when a child presses on different areas of the books' pages. The housing also contains a processor, memory, and a speaker to produce sounds. In one mode of operation, the SSR allows the child to press the first letter of a word and hear the sound of that letter.

The remainder of the letters in the word are grouped together and played together. For example, the child can press the letter “t” and hear the t phoneme and then press “ug” to hear all the sounds in the word “tug.” Similarly, the child can press the letter “b” and then “ug” to hear the sounds in “bug.” The SSR does not include a reader that allows the processor to automatically identify the inserted book. Instead, the user can press a triangle printed on the first page of the book, and the processor determines from the location of the triangle printed on the page which book is inserted. Similarly, the user can press a star on each page of the book, and the processor determines from the location of the star on the page which page of the book is being viewed. Thus, the SSR provides a roadmap for one of ordinary skill in the art desiring to produce an electronics-based learning toy for children that allows the use of phonetic-based learning methods, including the association of individual letters with their phonemes.

We agree with the district court that one of ordinary skill in the art of children's learning toys would have found it obvious to combine the Bevan device with the SSR to update it using modern electronic components in order to gain the commonly understood benefits of such adaptation, such as decreased size, increased reliability, simplified operation, and reduced cost. While the SSR only permits generation of a sound corresponding to the first letter of a word, it does so using electronic means. The combination is thus the adaptation of an old idea or invention (Bevan) using newer technology that is commonly available and understood in the art (the SSR). We therefore also find no clear error in the finding of the district court that one of ordinary skill in the art could have utilized the electronics of the SSR device, with the method of

operation taught by Bevan, to allow a child to press each individual letter in a word and hear the individual phonemes associated with each letter to sound out the words.

This combination of Bevan and the SSR lacks only the “reader” of claim 25 of the ’861 patent. The district court found that readers were well-known in the art at the time of the invention. As there is ample evidence in the record to support that finding, we find no clear error in the court’s determination. Furthermore, the reasons for adding a reader to the Bevan/SSR combination are the same as those for using readers in other children’s toys—namely, providing an added benefit and simplified use of the toy for the child in order to increase its marketability. Leapfrog presents no evidence that the inclusion of a reader in this type of device was uniquely challenging or difficult for one of ordinary skill in the art. See KSR, 2007 WL 1237837, at *15. Nor does Leapfrog present any evidence that the inclusion of a device commonly used in the field of electronics (a reader), and even in the narrower art of electronic children’s toys, represented an unobvious step over the prior art. Our conclusion is further reinforced by testimony from the sole inventor at trial that he did not have a technical background, could not have actually built the prototype himself, and relied on the assistance of an electrical engineer and Sandia National Laboratory to build a prototype of his invention.

Finally, we do not agree with Leapfrog that the court failed to give proper consideration to secondary considerations. The district court explicitly stated in its opinion that Leapfrog had provided substantial evidence of commercial success, praise, and long-felt need, but that, given the strength of the prima facie obviousness showing, the evidence on secondary considerations was inadequate to overcome a final

conclusion that claim 25 would have been obvious. We have no basis to disagree with the district court's conclusion.

In light of our review of the evidence and the lack of any clear error in the district court's factual findings, we agree with the district court's conclusion that claim 25 of the '861 is invalid as obvious in view of the combination of Bevan, the SSR device, and the knowledge of one of ordinary skill in the art concerning readers.

CONCLUSION

For the reasons stated, we affirm the district court's grant of judgment that Fisher-Price's PowerTouch device does not infringe claim 25 of the '861 patent and that claim 25 of the '861 patent is invalid as obvious.

AFFIRMED

The storage life of beef and pork packaged in an atmosphere with low carbon monoxide and high carbon dioxide

Oddvin Sørheim^{a,*}, Hilde Nissen^a, Truls Nesbakken^b

^aMATFORSK-Norwegian Food Research Institute, Osloveien 1, N-1430 Ås, Norway

^bNorwegian Meat Cooperative, Department of Research and Development, PO Box 360 Økern, N-0513 Oslo, Norway

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Abstract

Ground beef, beef loin steaks and pork chops were packaged in modified atmospheres of 0.4% CO/60% CO₂/40% N₂ and 70% O₂/30% CO₂. In addition ground beef was packaged in clipped chub packs, beef loin steaks were vacuum packaged, and pork chops were packaged in an atmosphere of 60% CO₂/40% N₂ with each pack containing an O₂ absorber. The packs were stored in the dark at 4 or 8°C for up to 21 days. Meat in 0.4% CO/60% CO₂/40% N₂ had a stable bright red colour that lasted beyond the time of spoilage. The storage lives in this gas mixture at 4°C, as limited by off-odours, were 11, 14 and 21 days for ground beef, beef loin steaks and pork chops, respectively. The 70% O₂/30% CO₂ atmosphere resulted in an initially bright red to red colour of the meat, but the colour was unstable and off-odours developed rapidly. The off-odours probably were caused by *Brochothrix thermosphacta*, which grew in all meat types, or by pseudomonads in ground beef. Meat stored in chub packs, vacuum packs or 60% CO₂/40% N₂ with an O₂ absorber developed off-odours and microflora similar to those of meat in 0.4% CO/60% CO₂/40% N₂, but with less acceptable appearances. These results show that a low CO/high CO₂ atmosphere is effective for preserving retail-ready meat. © 1999 Elsevier Science Ltd. All rights reserved.

1. Introduction

The main reasons for modified atmosphere packaging (MAP) of red meats for retail sale are to prolong the microbiological shelf life and to maintain an attractive red colour of the product. Modified atmospheres (MA) usually consist of carbon dioxide (CO₂) for inhibiting microbiological growth, oxygen (O₂) for enhancing colour and, occasionally, nitrogen (N₂) as a filler. The most common gas mixture for retail-ready meat contains approximately 70% O₂ and 30% CO₂, and gives the product an extended shelf life compared to air (Gill, 1996). The shelf life and colour stability of meat stored in this gas mixture is still limited. To obtain a stable red colour for the meat, low concentrations (<1%) of carbon monoxide (CO) can be introduced in the MA. Then, O₂ can be removed from the gas mixture and the concentration of bacteriostatic CO₂ can be increased. Anaerobic conditions extend the shelf life of meat considerably compared to air and O₂-enriched atmospheres (Gill & Molin, 1991). CO binds strongly to the meat

pigment myoglobin to form stable carboxymyoglobin which has a cherry red colour (El-Badawi, Cain, Samuels, & Angelmeier, 1964). Low concentrations of CO have little effect on the microflora of meat (Clark, Lentz, & Roth, 1976; Gee & Brown, 1978; Luño, Beltrán, & Roncalés, 1998).

The Norwegian meat industry has for the past decade been using a gas mixture of approximately 0.3–0.5% CO, 60–70% CO₂ and 30–40% N₂ in retail-ready packages of beef, pork and lamb. Packages with this gas mixture now have a 50–60% share of the domestic, retail, red meat market. The technological, hygienic and toxicological aspects of using CO in MA for meat have recently been reviewed with the conclusion that CO used in concentrations up to 1% does not present a toxic hazard to the consumer (Sørheim, Aune, & Nesbakken, 1997a). However, CO may mask spoilage, because the stable cherry red colour can last beyond the microbiological shelf life of the meat (Kroof, 1980).

The inclusion of CO in MA for meat is controversial. CO is presently not allowed in MA for meat in the USA and in the EU (Cornforth, 1994; European Parliament and Council Directive, 1995). However, Norwegian food control authorities have up to now not opposed

* Corresponding author. Tel.: +47-64-970100; Fax: +47-64-970333; E-mail: oddvin.sorheim@matforsk.no

the use of up to 0.5% CO in MA for meat. This would change with an adoption of EU food regulations in Norway. Consequently, the Norwegian meat industry is seeking amendments of current EU food regulations relating to the use of CO in MAP of red meats. If the use of CO should be disallowed, other means of maintaining the long shelf life and the attractive red colour of the meat will have to be sought.

The aim of the present experiments was to compare a commercial Norwegian CO/CO₂/N₂ mixture with alternative gas mixtures and packaging methods for their effects on the off-odour, microflora and colour of ground beef, beef loin steaks and pork chops stored at 4 or 8°C for up to 21 days.

2. Materials and methods

2.1. Preparation of meat

2.1.1. Ground beef

Twenty cow and bull carcasses of Norwegian Red Cattle, which weighed on average 275 kg, were electrically stimulated with 90 V and were chilled using programmed air temperatures between 12 and –5°C. Two days after slaughter the carcasses were deboned, and trimmings with 14% fat were ground through a 4 mm plate. The batch of ground beef was divided into 500 g portions.

2.1.2. Beef loin steaks

Loins (*m. longissimus lumborum et thoracis*) with ultimate pH values below 5.8 were deboned from 25 bull carcasses of Norwegian Red Cattle. These carcasses, which weighed on average 275 kg, were stimulated, chilled and deboned the same way as the carcasses used in the preparation of ground beef. The loins were vacuum packaged and aged for 11 days at 3°C. Thereafter, the loins were cut into steaks 2.5 cm thick, and were randomly assigned to retail packs which each contained two steaks.

2.1.3. Pork chops

Thirty pig carcasses of Norwegian Land Race, which weighed on average 75 kg, were blast-chilled. Four days after slaughter, bone-in loins were removed and crust-frozen in liquid N₂ at –50°C for 20 min to facilitate cutting of chops. The chops, which were 1.6 cm thick, were randomly assigned to retail packs which each contained two chops.

2.2. Packaging

Ground beef, beef loin steaks and pork chops were packaged in 0.4% CO/60% CO₂/40% N₂ (CO mixture) and 70% O₂/30% CO₂ (high O₂). In addition, ground beef was packaged in clipped chub packs, beef loin steaks were vacuum packaged and pork chops were packaged in 60% CO₂/40% N₂ with one Ageless® FX-

100 O₂ absorber (Mitsubishi Gas Chem. Co. Inc., Tokyo, Japan) in each pack (mixture with O₂ absorber).

The meat was packaged at a commercial meat plant within 2 h of grinding or cutting. Meat in the CO mixture, the high O₂ mixture and the mixture with O₂ absorber was packaged in an Ilapak Delta 2000 flow-packaging machine (Ilapak Machine Auto S.A., Grancia, Switzerland). The CO mixture was a blend of 1% CO/99% N₂ with 100% CO₂. The high O₂ mixture was used as a preblend. The mixture with O₂ absorber was a blend of 100% N₂ with 100% CO₂ (all gases, Hydrogas, Porsgrunn, Norway). The initial gas volume to meat weight ratio in the packs was approximately 1.5 to 1. The packs consisted of polyethylene trays (Færch Plast, Holstebro, Denmark) wrapped in Cryovac BDF 550 shrinking film (Cryovac, Milan, Italy) with an O₂ transmission rate of 19 cm³/m²/24 h/atm at 23°C and 0% RH. Chub packs of ground beef were packaged in a clipping machine (Poly-Clip, Frankfurt, Germany) using a red, fishingnet-patterned, polyethylene film (SFK, Vidovre, Denmark) with an O₂ transmission rate of 500 cm³/m²/24 h/atm at 23°C and 0% RH. Beef loin steaks were vacuum packaged in a Multivac 5100 thermo-forming machine (Multivac, Wolfertschwenden, Germany) using a terephthalate/polyethylene upper film and polyamide/polyethylene lower film with O₂ transmission rates of 10 and 16 cm³/m²/24 h/atm at 23°C and 0% RH, respectively (Danisco, Horsens, Denmark).

2.3. Storage and sampling of meat

Five samples were collected from the ground beef batch, beef loins and pork loins before packaging, for pH measurements and microbiological analyses.

The packaged meat was stored in dark chilling rooms at 4 ± 0.5 or 8 ± 0.5°C for up to 21 days at least until off-odours developed. Five packs were removed per product, packaging method, storage temperature and sampling day after the following storage times:

- ground beef: 2, 4, 6, 8 or 11 days;
- beef loin steaks: 3, 7, 10 or 14 days; and
- pork chops: 3, 7, 10, 14, 17 or 21 days.

2.4. Gas analyses

The atmospheres of packs with MA were analysed for O₂ and CO₂ immediately after packaging (approximately every tenth pack) and at sampling (all packs). O₂ was determined using a Toray LC 700-F gas analyser (Toray Engineering, Osaka, Japan) and CO₂ using a Toray PG-100 gas analyser (Toray). The threshold levels for the O₂ and CO₂ analyses were 0.05 and 1%, respectively. Gas samples of 10 cm³ were removed with a syringe through selfsealing patches on the packs.

2.5. pH

The pH measurements were made directly in the meat with an Ingold Xerolyt gel electrode (Mettler-Toledo A.G., Greifensee, Switzerland).

2.6. Odour

The meat was evaluated for odours by a three member trained panel between 0.5 and 1 min after opening of the packs. The off-odour scale used was: 1 = none, 3 = slight and 5 = extreme. Scores of 3 or below were considered acceptable.

2.7. Microbiology

Ten gram meat samples were collected from portions of the ground beef, and diluted in 90 g peptone water. A sample 25 cm² and 2–3 mm thick was removed from the surface of each beef loin or steak and pork loin or chop with a scalpel, and diluted in 100 ml peptone water. Each sample was macerated in a Stomacher for 1 min. Serial 10-fold dilutions of each Stomacher fluid were prepared, and 20 µl volumes of appropriate dilutions were plated in duplicate on the following media:

- plate count agar (PCA; Difco, Difco Laboratories, Detroit, MI, USA) for total viable counts;
- de Man, Sharpe and Rogosa agar (MRS; Oxoid, Unipath Ltd., Basingstoke, Hampshire, UK) adjusted to pH 5.7 for lactic acid bacteria (de Man, Rogosa, & Sharpe, 1960);
- streptomycin thallous acetate actidione agar base (STAA; CM 881 with selective supplement SR 151; Oxoid) for *Brochothrix thermosphacta*;
- pseudomonads agar base (CFC; CM 559 with selective supplement SR 103; Oxoid) for pseudomonads;

In addition, 1 ml portions of appropriate dilutions were plated in duplicate on petrifilm coliform count plates (3M Microbiology Products, St. Paul, MN, USA) for enumeration of coliforms and *Escherichia coli*.

Plates of PCA, MRS, STAA and CFC were incubated at 20°C for four days, and petrifilm plates at 30°C for up to 2 days, all aerobically. Counts were expressed as colony forming units (CFU) per g or cm².

2.8. Colour

A six-member trained panel evaluated the colour of the meat in intact packs under 1200 ± 200 lux Warnton Lumilux L36W/31 yellow-white light (Osram, Drammen, Norway). The colour was assessed on a scale where 1 = bright red (ground beef and beef loin steaks) or light bright red (pork chops), 2 = red (ground beef

and beef loin steaks) or light red (pork chops), 3 = slightly brown, grey or green, 4 = moderately brown, grey or green and 5 = extremely brown, grey or green (National Live Stock and Meat Board, 1991).

A Minolta Chroma Meter CR-300 (Minolta Camera Co., Osaka, Japan) with 8 mm viewing port and illuminant D₆₅ was used for measuring CIE *a** values (redness). The colour was measured directly at the meat surface within 1 min of opening of each pack.

Ground beef in chub packs was not included in the colour analyses because the red packaging film hides the colour of the product. With pork chops, the colour of only the *m. longissimus lumborum et thoracis* was analysed.

2.9. Statistics

Analysis of variance by Tukey's multiple comparisons test was performed using the Systat programme, version 6 (Systat Inc., Evanston, IL, USA).

3. Results

3.1. Gas composition

The initial O₂ concentrations in packs with the CO mixture and the mixture with O₂ absorber were all below 0.5% immediately after packaging. O₂ was not detected in these packs after 2 or 3 days storage. The level of O₂ in packs of high O₂ was reduced from the initial 70 to 60–65% during storage for up to 21 days. Concentrations of CO₂ in the packs were generally reduced by one fifth after 2 or 3 days storage, and were then stable (data not shown).

3.2. Storage life of ground beef

The time to develop off-odours was 2 to 3 days longer for ground beef stored in the CO mixture and in chub packs than in high O₂, and it was 4 or 5 days longer at 4 than at 8°C for all three packaging methods (Table 1). In high O₂, the total viable counts increased faster and were higher (*p* < 0.01) than for the other two types of packaging after 2 days at either 4 or 8°C [Fig. 1(a)]. The total viable counts were more than 90% lactic acid bacteria (data not shown). The high numbers of lactic acid bacteria in ground beef, up to approximately log₁₀ 8 CFU/g, caused a decrease in the pH value from the initial 5.7 to 5.2 after 6 days when the meat was stored in the CO mixture or chub packs at 8°C (data not shown). At 4°C, the pH value was reduced to 5.5 after 11 days in both those packaging systems. The numbers of *B. thermosphacta* increased, in meat in high O₂ [Fig. 1(b)]. In meat in high O₂ the numbers of pseudomonads increased up to approximately log₁₀ 7 CFU/g, but only to log₁₀ 5 and 6 CFU/g in

meat in the CO mixture or chub packs, respectively (data not shown).

Ground beef in the CO mixture had a stable bright red colour, as shown by both the low colour scores and the high a^* values [Fig. 1(c) and (d)]. Meat in high O_2 was significantly less red ($p < 0.05$) than meat in the CO mixture, with higher colour scores and lower a^* values at day 2 and at later storage times at both 4 and 8°C. The colour of meat in high O_2 deteriorated with time, significantly faster ($p < 0.01$) at 8 than at 4°C.

Table 1
Time for development of off-odours in different types of meat in various packagings at storage temperatures of 4 or 8°C

Product	Packaging ^a	Time of off-odour detection (days)	
		4°C	8°C
Ground beef	CO mixture	11	6
	High O_2	8	4
	Chub packs	11	6
Beef loin steaks	CO mixture	14	7
	High O_2	10	7
	Vacuum packs	14	7
Pork chops	CO mixture	21	14
	High O_2	14	7
	Mixture with O_2 absorber	17	10

^a CO mixture = modified atmosphere of 0.4% CO/60% CO₂/40% N₂; High O_2 = modified atmosphere of 70% O₂/30% CO₂; Mixture with O_2 absorber = modified atmosphere of 60% CO₂/40% N₂ with an O_2 absorber in the pack.

3.3. Storage life of beef loin steaks

At 4°C, off-odours developed 4 days later in beef loin steaks in the CO mixture and in vacuum packs than in high O_2 (Table 1). At 8°C, no differences in the development of off-odours were observed. Off-odours developed 4 to 7 days earlier in meat at 8 than at 4°C. The type of packaging did not significantly affect ($p < 0.05$) the total viable counts on the meat, but the counts were significantly higher ($p < 0.01$) at 8 than at 4°C after both 3 and 7 days of storage [Fig. 2(a)]. The numbers of *B. thermosphacta* were less than log₁₀ 4 CFU/cm² in meat in all types of packaging at all times, but were significantly higher ($p < 0.05$) on meat in high O_2 at 7 and 10 days than on meat in the CO mixture and in vacuum packs at equivalent times [Fig. 2(b)]. The numbers of pseudomonads did not exceed log₁₀ 3.5 CFU/cm² at any sampling time, and were not significantly affected ($p > 0.05$) by the type of packaging or the storage temperature.

The colour of the beef loin steaks in the CO mixture was stable bright red throughout storage at both 4 and 8°C, as shown by the low colour scores and high a^* values [Fig. 2(c) and (d)]. Steaks in high O_2 were also bright red with high a^* values at day 3, but these steaks discoloured gradually between days 3 and 10, significantly faster ($p < 0.05$) at 8 than at 4°C. Meat in vacuum packs was slightly discoloured with low a^* values throughout storage. The colour scores and a^* values of vacuum packaged steaks were not significantly affected ($p > 0.05$) by the storage temperature.

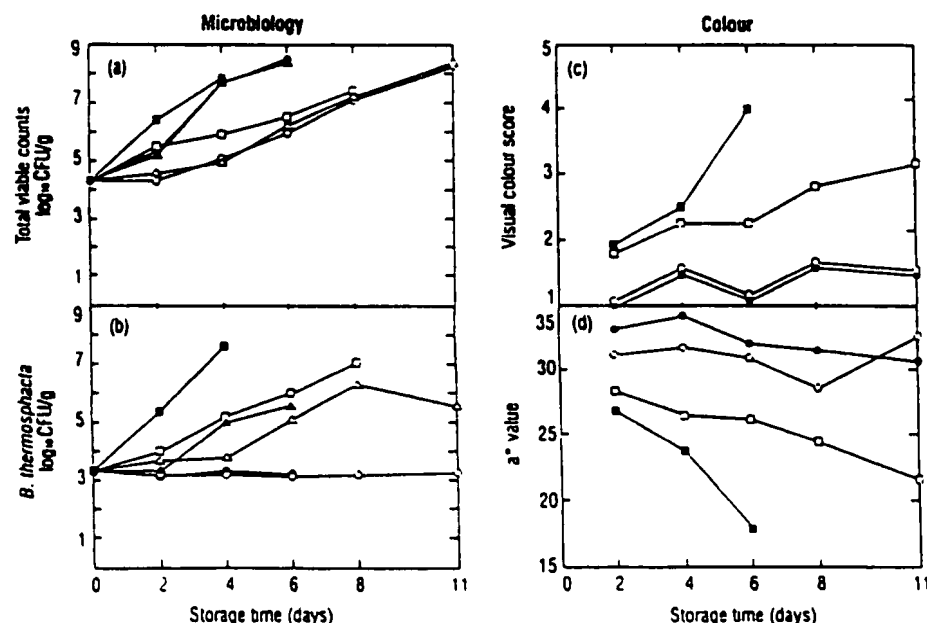


Fig. 1. Mean values ($n = 5$) for (a) total viable counts, (b) numbers of *Brochothrix thermosphacta*, (c) visual colour scores and (d) CIE a^* values for ground beef stored in 0.4% CO/60% CO₂/40% N₂ at 4°C (○) or 8°C (●), in 70% O₂/30% CO₂ at 4°C (□) or 8°C (■), or in chub packs at 4°C (△) or 8°C (▲). Colour was assessed on a scale where 1 = bright red and 5 = extremely discoloured.

3.4. Storage life of pork chops

For pork chops, off-odours developed more slowly in meat in the CO mixture than in meat in the mixture with O₂ absorbers or in high O₂ (Table 1). Off-odours were detected 7 days earlier at 8 than at 4°C for chops in each type of packaging. The type of packaging did not affect the total viable counts on the pork chops [Fig. 3(a)]. However, the counts were greater on meat stored at 8 than at 4°C. The numbers of *B. thermosphacta* on chops in high

O₂ were significantly higher ($p < 0.01$) than on chops in the CO mixture or in the mixture with O₂ absorbers after 7 days at 8°C or 10 days at 4°C, and reached approximately $\log_{10} 6$ CFU/cm² [Fig. 3(b)]. The numbers of pseudomonads did not exceed $\log_{10} 3$ CFU/cm² on any of the pork chops.

The colour of pork chops in the CO mixture was light bright red with high *a** values throughout storage [Fig. 3(c) and (d)]. Chops in high O₂ were red at day 3, but discoloured during storage, significantly faster ($p < 0.05$) at

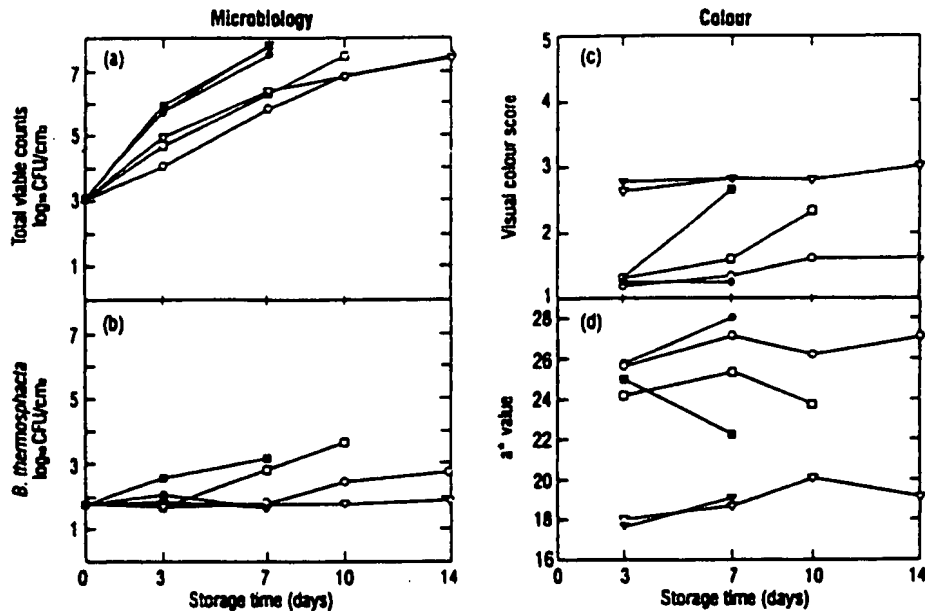


Fig. 2. Mean values ($n = 5$) for (a) total viable counts, (b) numbers of *Brochothrix thermosphacta*, (c) visual colour scores and (d) CIE *a** values for beef loin steaks stored in 0.4% CO/60% CO₂/40% N₂ at 4°C (○) or 8°C (●), in 70% O₂/30% CO₂ at 4°C (□) or 8°C (■), or in vacuum packs at 4°C (▽) or 8°C (▼). Colour was assessed on a scale where 1 = bright red and 5 = extremely discoloured.

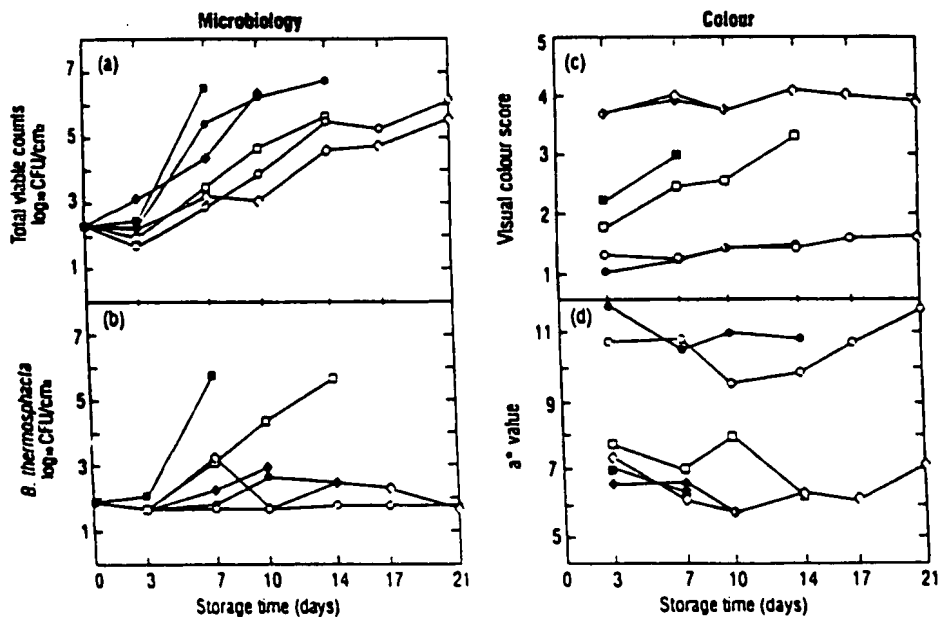


Fig. 3. Mean values ($n = 5$) for (a) total viable counts, (b) numbers of *Brochothrix thermosphacta*, (c) visual colour scores and (d) CIE *a** values for pork chops stored in 0.4% CO/60% CO₂/40% N₂ at 4°C (○) or 8°C (●), in 70% O₂/30% CO₂ at 4°C (□) or 8°C (■), or in 60% CO₂/40% N₂ with O₂ absorbers at 4°C (○) or 8°C (◆). Colour was assessed on a scale where 1 = light bright red and 5 = extremely discoloured.

8 than at 4°C. Approximately 75% of the chops in high O₂ had black back bones at the time of sampling. Chops in the mixture with O₂ absorbers were moderately discoloured from day 3 to the end of storage. These chops had *a** values similar to those of chops in high O₂.

4. Discussion

4.1. Off-odour and microflora

The shelf life of the meat, as determined by the time to develop off-odours, was influenced by the packaging method, the storage temperature and the initial microbiological load on the meat. Storage of meat in the CO mixture, in vacuum packs or in chub packs gave the longest shelf lives. Meat stored in high O₂ generally developed off-odours 2-7 days earlier at 4 or 8°C than meat packaged in the other gas mixtures or by the other methods.

The differences in the rates of development of off-odours, as affected by the packaging method, were seldom related to any differences in numbers of total viable counts. However, the development of off-odours from the three meat types, especially ground beef and pork chops in high O₂, coincided with the attainment of high numbers of *B. thermosphacta*. For ground beef, storage in the CO mixture retarded growth of *B. thermosphacta* even more than storage in chub packs. At chill temperatures above 1°C, *B. thermosphacta* often causes spoilage of meat stored in high O₂ atmospheres (Dainty & Mackey, 1992). High concentrations of CO₂, removal of O₂ and low storage temperature inhibit the growth of *B. thermosphacta* (Gill, 1996; Nissen, Sørheim, & Dainty, 1996). Pseudomonads probably contributed to the off-odours of ground beef. Meat in high O₂ is often spoiled by *Pseudomonas* spp., but the growth of pseudomonads is retarded under anaerobic conditions (Dainty & Mackey, 1992; Gill, 1996). A shift in the metabolism of lactic acid bacteria under aerobic conditions can also produce off-odours (Nissen et al., 1996). In the present experiments, the numbers of coliforms or *E. coli* did not exceed log₁₀ 3 CFU/g or cm² in any samples. Therefore, those organisms probably did not contribute to off-odours.

For pork chops, the effect of CO on the microflora can be evaluated because the gas compositions of the CO mixture and of the mixture with O₂ absorber were identical, except for the inclusion of 0.4% CO in the former. Although a 4 day increase in the time to develop off-odours was observed with the CO mixture, there was no significant reduction in the microbiological counts. Luño et al. (1998) used 1% CO in high O₂ atmospheres and noted a delay in the onset of off-odours without any reduction in the numbers of psychrotrophic bacteria. However, Clark et al. (1976) found that the addition of

0.5-10% CO to N₂ atmospheres reduced the number of psychrotrophic bacteria and increased the odour shelf life of beef. For example, 1.0% CO in 99% N₂ increased the time to develop off-odours at 5°C from 18 to 24 days. The lack of such an effect of CO on bacteria in our experiments may be due to the use of 60% CO₂ overshadowing any effect of CO.

The use of CO makes it possible to dispense with O₂ and so to increase the CO₂ concentration in a MA to about 60%. Our data suggest that 0.4% CO probably has little or no direct effect on the growth of bacteria. Other studies have shown that increasing the CO₂ concentration from 20 to 100% increases the bacteriostatic effect of the gas, but the efficiency is highly dependent on low storage temperatures (Gill & Molin, 1991; Nissen et al., 1996). The high CO₂ concentration and absence of O₂ in the CO mixture will favour the growth of lactic acid bacteria, which usually cause a mild form of spoilage only late in the development of the spoilage flora (Gill, 1996).

The present experiments were performed at acceptable and abusive storage temperatures to assess the effects of temperatures commonly encountered in the distribution and sale of retail-ready meat. The storage temperature strongly affected the rates of growth of microflora and the time to develop off-odours. Consequently, independently of the packaging method, the shelf life of meat can be considerably extended by maintaining low temperatures in the chill chain (Gill & Molin, 1991; Nissen et al., 1996).

4.2. Colour

The CO mixture gave a stable bright or light bright red colour with consistent high *a** values for all three products, irrespective of the storage temperature. The initial level of residual O₂, up to 0.5%, did not adversely affect the visual scores and instrumental values for the colour of meat stored in the CO mixture.

CO binds to myoglobin and forms cherry red carboxymyoglobin (El-Badawi et al., 1964). This pigment is spectrally similar to the bright red oxymyoglobin which normally develops at the surface of fresh meat in air. Carboxymyoglobin is less readily oxidized to brown metmyoglobin than is oxymyoglobin, because of the strong binding of CO to the iron-porphyrin site on the myoglobin molecule (Lanier, Carpenter, Toledo, & Reagan, 1978; Wolfe, 1980). Consequently, CO in concentrations of 0.5-2.0% enhances and stabilizes a bright red colour of meat (Kropf, 1980; Sørheim et al., 1997a). In a recent study, 1% CO in combination with 24 or 70% O₂ stabilized the colour of beef by reduced formation of metmyoglobin after storage at 1°C for up to 29 days (Luño et al., 1998). However, in a study of beef stored in a MA of 2% CO/78% CO₂/20% N₂, the colour of the meat was characterized as "too artificial" by

a sensory panel (Renner & Labadie, 1993). From our studies and experience from the Norwegian meat industry, 0.4% CO seems sufficient to produce a stable, attractive, bright red colour of meat.

All three meat types stored in high O₂ were bright red to red with high *a** values early in the storage periods, approaching the colour of meat in the CO mixture. As the microbiological counts of meat in high O₂ increased, the colour deteriorated, faster at 8 than at 4°C. Meat stored in a MA of high O₂ develops a thicker layer of oxymyoglobin than meat stored in air (Renner & Labadie, 1993). However, the oxymyoglobin gradually oxidizes to metmyoglobin, and the oxidation is faster at higher temperatures.

For cut bone, haemoglobin released from disrupted red blood cells in the marrow will accumulate at the surface and ultimately become black after the bone has been exposed to air or O₂ (Gill, 1996). Although bone blackening was not considered in the present visual colour evaluation, it can negatively affect the saleability of bone-in meat at retail display. The cut bones of pork chops stored in high O₂ blackened during storage, but this discoloration was not observed on bones in the CO mixture and the mixture with O₂ absorbers.

Beef loin steaks stored in vacuum packs were slightly discoloured with low *a** values at both 4 and 8°C. In these packs, meat juices were observed between the upper and lower films, but that did not influence the colour evaluations.

O₂ absorbers in packs with high CO₂ facilitate the removal of residual O₂ and maintain atmospheres free of O₂ during storage (Smith, Abe, & Hoshino, 1995). Low levels of residual O₂, above 0.01–0.15% for beef and 0.5–1.0% for pork, will inevitably discolour the meat (Penney & Bell, 1993; Gill, 1996; Sørheim et al., 1997b). When no CO is present in an O₂ depleted MA, it is essential to remove the residual O₂ as fast and completely as possible to avoid formation of metmyoglobin. In these experiments, pork chops stored in the gas mixture with O₂ absorbers were moderately discoloured during the whole storage period at 4 or 8°C. Despite the obvious visible differences, these chops had similar *a** values to the chops in high O₂. The discoloured surface made the chops unfit for sale, even in the early stage of storage. The present findings contrast with previous results, where the colour of porcine *m. longissimus thoracis et lumborum* was significantly improved by using O₂ absorbers in MAs of CO₂ with residual O₂ (Sørheim et al., 1997b). The present discoloration could be caused by incomplete use or function of the absorbers (Gill, 1996).

4.3. Benefits and disadvantages of a MA with low CO/high CO₂

An objection raised against using CO as a small component of a MA for retail-ready meat is the possi-

bility that the colour stability can exceed the microbiological shelf life, with the risk of masking spoilage of the meat (Kropf, 1980). Therefore, the consumer must evaluate the microbiological condition of meat in a CO mixture by off-odours. When a MA with CO is applied commercially, it is important to have a proper control of the hygienic condition of the meat raw materials and the chill chain temperatures.

CO used in concentrations below 1.0% does not present any hazard to the consumer, because consumption of meat packaged in such concentrations of CO will result in only negligible levels of carboxyhaemoglobin in the blood of consumers (Sørheim et al., 1997a). By delivering CO in a 1% mixture with 99% N₂, which is the practice of Norwegian gas suppliers, CO is considered safe for use in the working environment. Other MAs with high levels of O₂, up to 70%, must be regarded as explosive gas mixtures, which must be used with appropriate precautions for safety (Luño et al., 1998).

The suitability of gas mixtures and packaging methods for red meats for retail display depends on their ability to both reduce spoilage and stabilize colour. Gas mixtures with low concentrations of CO and high concentrations of CO₂ provide a combination of a long microbiological shelf life and a stable, bright red colour of meat. Meat packaged in a MA with high O₂ can achieve an initial bright red colour, but the microbiological shelf life and the colour stability are both considerably lower than those of meat in the CO mixture. Using CO eliminates the need to have O₂ as a component of the MA. Other MAs and packaging methods, like high CO₂ with O₂ absorbers, chub packs and vacuum packs may give a shelf life comparable to that of the CO mixture, but with a less acceptable colour or appearance of the meat. Thus, there appears at present to be no fully satisfactory alternative to the CO mixture used in packaging of retail-ready red meats in Norway.

Acknowledgements

The financial support of this study from the Research Council of Norway is highly appreciated. Vestfold-Buskerud Slakteri A/L, Sem and Hydrogas AS Utviklings-senter, Porsgrunn, are greatly thanked for packaging of the meat. We appreciate the gift of Ageless® O₂ absorbers from Cryovac Europe, Norderstedt, Germany. The technical staff and Per Lea (statistics) at MATFORSK are thanked for their skilful assistance in the study.

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~~CONFIDENTIAL~~
MEMORANDUM OF CONFERENCE

May 12, 1962

BETWEEN: Mr. Donald W. Thomas, Legal Counsel, The Whirlpool Corporation
Muskegon Harbor, Michigan

and

Mr. A. T. Spilner, Jr., Food Additive Petitions Control Branch

SUBJECT: Combustion product gas.
Food Additive Petition 731.

Mr. Thomas called without previous appointment to discuss the above petition. He said that he had received my letter of May 10, 1962, in which we filed the petition, and said that we may need additional data on meat. These data would be needed to establish that the treatment of meat would not serve to cause the meat to retain its fresh red color longer than meat not so treated.

I explained to Mr. Thomas the way in which petitions are handled, and explained the question which we have concerning possible deception of the consumer where treatment of the meat leads to longer retention of the fresh red color. I said that they could either submit additional data on this point or they could request withdrawal of the portion of the petition for meat, and explained the different courses of action.

Mr. Thomas said that they had data concerning the retention of red color in meat, and they will get it together. He was concerned, however, about whether he should submit this as an amendment which would start the time clock over, or should withdraw animal products from the petition, to submit later on.

I said that this was a decision which he would have to make in the light of the explanation we had given him, and I suggested that he submit the data which they have and let us look at it before they did anything additional, because what they had done might be sufficient for our people.

I further suggested that when he submit the information for meat, he should supplement the data in the petition to explain exactly how the combustion product gas is to be used on the various commodities named in their petition. He said that he would do so. Briefly, he said that the gas was to be used as the atmosphere in a cold storage room.

In response to a question, he said that they had tested the effluent from their generator and were satisfied that the gas complied with the requirements established in the food additive regulation.

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32° and 45° F. and in their normal distribution containers, that is, baskets, crates and boxes. Products so stored had a shelf life of from three to five times that of air-stored food held at the same temperature. The results of one such study involving apples stored in air versus apples stored in conventional controlled atmosphere versus apples stored in combustion product gas are presented in the attached table. It will be noted that apples stored in combustion product gas had firmer flesh and a lower incidence of scald than did apples stored either in air or conventional controlled atmosphere even though the apples in combustion product gas were in storage for a longer period of time.

The combustion product gas under study at Whirlpool would most likely be used in the following general areas:

1. Fresh fruit and vegetable storage
2. Processors - storage, packaging and processing
3. Transportation

Because of these diverse applications, our petition requests approval for fruit and vegetable "products" as well as the natural, original raw fruits and vegetables.

To expand on the use of combustion product gas by food processors, the following examples are presented:

1. Storage of fruits and vegetables in order to have better quality control, improve yield and extend packaging season.
2. Packaging of processed foods in inert gases, i. e., nitrogen and/or carbon dioxide to prevent oxidative changes that may develop during storage.
3. Use of gas mixtures in certain processing steps as a "blanket" to keep out oxygen and prevent the associated undesirable changes.

Mr. Alan T. Spiher, Jr.

Page Three

We are hopeful that the requested amendment to the petition as well as the supplemental information presented above will clear up any questions concerning Food Additive Petition No. 751 and that favorable action will be shortly forthcoming.

Very truly yours,

WHIRLPOOL CORPORATION

By William E. Mahaffey
Vice President

KALSEC

November 15, 2005



Division of Dockets Management (HFA-305)
Food and Drug Administration
Department of Health and Human Services
Room 1061
5630 Fishers Lane
Rockville, MD 20852

CITIZEN PETITION

A. Action Requested

This Citizen Petition is submitted by Kalsec, Inc. ("Kalsec") under Sections 201, 402, 403, 409, and 721 of the Food, Drug, and Cosmetic Act ("FDCA" or "the Act") and Section 10.30 of the Food and Drug Administration's ("FDA") implementing regulations. Kalsec produces spice, herb, hop, and vegetable extracts for use in food, beverage, and pharmaceutical applications. By this Citizen's Petition, Kalsec requests that FDA take immediate action to prohibit the use of carbon monoxide in the packaging of fresh meat, including to terminate the agency's unlawful responses to the Generally Recognized As Safe ("GRAS") notifications submitted by Pactiv Corp. and Precept Foods, Inc., GRAS Notice Nos. GRN 000083 and 000143 ("GRN 83" and "GRN 143"), and taking all such further actions as are necessary to effectively implement and enforce an immediate ban on carbon monoxide in fresh meat packaging, in coordination with USDA Food Safety and Inspection Service ("FSIS"). Kalsec advocates the actions requested to prevent serious harms to public health and consumer confidence in the integrity of the U.S. meat supply.¹

B. Statement of Grounds

1. The Pactiv and Precept GRAS Notifications

FDA has failed to object to GRAS notifications for the unlawful use of carbon monoxide to impart color to fresh meat products. On February 21, 2002, FDA responded to a

¹ It is well established that carbon monoxide has effects on the color of fresh meat. *See, e.g.*, scientific literature cited at note 91, *infra*, and attached as Attachments 16-18; *see also* "Pathogen Inoculation Study of Ground Beef Under Modified Atmosphere Package (MAP) Conditions," S&J Laboratories, Inc. (November 14, 2005), examining the effects of carbon monoxide on the color of fresh meat under a variety of laboratory conditions (Attachment 1).

Address:
P.O. Box 50511
Kalamazoo, MI 49005-0511

3713 West Main St
Kalamazoo, MI 49006

Telephone:
269-349-9711
800-323-9320

Web:
www.kalsec.com

Fax:
Sales & Marketing 269-382-3060
Hop Sales & Lab 269-349-9055
Customer Service 269-349-1195
Purchasing 269-349-1558
Accounting 269-349-1558

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GRAS notification submitted on behalf of Pactiv Corporation ("Pactiv"),² informing FDA of its GRAS determination for the use of carbon monoxide gas, at levels of 0.4 percent, to displace oxygen inside packaging for fresh, case-ready red muscle meat and ground meat products.³ The FDA "no objection" letter expressly recognizes that the functional purpose of the carbon monoxide gas is to impart color to fresh meat, giving it "a desirable red color during storage."⁴

On July 29, 2004, FDA responded to a similar GRAS notification submitted on behalf of Precept Foods, LLC ("Precept")⁵ informing FDA of its GRAS determination for the use of carbon monoxide gas at levels of 0.4 percent to displace oxygen inside packaging for fresh, case-ready beef and pork products intended for direct sale to consumers. As in the case of the prior Pactiv notification, the FDA "no objection" letter again expressly recognizes that the functional purpose of the carbon monoxide is to impart color to fresh meat.⁶

In evaluating the GRAS notifications of Pactiv and Precept Foods, FDA consulted with the USDA FSIS under new FDA/USDA joint fast track premarket clearance procedures governing the approval of ingredients for meat products.⁷ FSIS subsequently issued "acceptability determinations" further implementing the unlawful allowance of carbon monoxide to impart color to fresh meat products,⁸ and FDA also continues to consider and allow expanded

² Letter from Alan M. Rulis, Director, CFSAN, Office of Food Additive Safety, to Eric Greenberg, Ungaretti and Harris (Feb. 21, 2002) ("Agency Response Letter to GRAS Notice No. GRN 000083"), available at <http://www.cfsan.fda.gov/~rdb/opa-g083.html>.

³ Under the conditions of use specified in the Pactiv GRAS notification, 0.4 percent carbon monoxide gas is blended together with 30 percent carbon dioxide and 69.6 percent nitrogen gases in the modified atmosphere packaging ("MAP") system. The case ready meats are intended to be removed from the MAP system prior to retail display. No labeling requirements are specified under these conditions of carbon monoxide use. Agency Response Letter to GRAS Notice No. GRN 000083, at 1.

⁴ *Id.* at 2.

⁵ Letter from Laura M. Tarantino, Director, Center for Food Safety and Applied Nutrition ("CFSAN"), Office of Food Additive Safety, to Gary J. Kushner and Anne M. Boekman, Hogan and Hartson (July 29, 2004) ("Agency Response Letter to GRAS Notice No. GRN 000143"), available at <http://www.cfsan.fda.gov/~rdb/opa-g143.html>.

⁶ *Id.* at 2.

⁷ See 65 Fed. Reg. 3330 (May 23, 2000); "Memorandum of Understanding Between The Food Safety and Inspection Service United States Department of Agriculture and The Food and Drug Administration United States Department of Health and Human Services Regarding the Listing or Approval of Food Ingredients and Sources of Radiation Used in the Production of Meat and Poultry Products," ("Meat Ingredients MOU") (Jan. 18-31, 2000), available at <http://www.fda.gov/OHRMS/DOCKETS/98fr/225002000.pdf>.

⁸ See FSIS Directive 7120.1, "Safe and Suitable Ingredients Used in the Production of Meat and Poultry Products," Amdt. 5 (October 13, 2005), listing FSIS acceptability determinations allowing two carbon monoxide packaging systems by Cryovac and two such systems by Cargill.

uses of carbon monoxide in fresh meat packaging based upon its improper responses to the Pactiv and Precept GRAS notifications.⁹

2. Summary of Argument

This Citizen Petition requests that FDA take immediate action to prohibit the use of carbon monoxide to displace oxygen in fresh meat packaging, including by withdrawing the agency's responses to the unlawful GRAS notifications submitted by Pactiv and Precept. The requested action is necessary to prevent serious harms to public health and consumer confidence in the safety and integrity of the U.S. meat supply. The requested ban of carbon monoxide in fresh meat packaging is required under FDCA provisions governing the use of color additives, food additives, and GRAS substances in food, and related provisions of the Federal Meat Inspection Act ("FMIA") governing the suitability of such ingredients in fresh meat products.¹⁰

The use of carbon monoxide in fresh meat packaging presents serious food safety and consumer deception concerns of the same kinds that historically justified the broad-based ban on color additives in fresh meat products. Carbon monoxide obscures the natural coloration of meat that is indicative of freshness and safety, by reacting with the natural myoglobin in meat to produce carboxymyoglobin, a bright red substance that hides the true colors of meat, simulating the appearance of freshness and masking meat spoilage. This color-masking effect is particularly dangerous in anaerobic packaging environments such as those described in the Pactiv and Precept GRAS notifications, which potentially allow the proliferation of pathogens such as *Clostridium botulinum* but inhibit the growth of aerobic spoilage organisms that provide the tell-tale signs of spoilage upon which consumers rely, in addition to color change, to determine that meat is no longer safe to consume. It is well established under the FDCA and FMIA that food ingredients are prohibited under conditions that are unsafe, conceal damage or inferiority, or make food appear better or of greater value than it is.¹¹

The color-imparting effects of carbon monoxide under the conditions of use in fresh meat packaging render the substance an unapproved and prohibited color additive. Neither FDA nor FSIS has the legal authority to permit the use of carbon monoxide in the packaging of fresh meat, in the absence of FDA regulations listing carbon monoxide under FDCA section 721. FSIS lacks the authority to make a suitability determination permitting the use of a color additive in meat, except where it has first been approved by FDA under FDCA section 721.¹² In the case of carbon monoxide, not only has FDA failed to issue the rules necessary to approve the use in fresh meat packaging, but the agency has also disregarded the explicit prohibition on this very use in fresh meat under its own food additive regulations.

⁹ See CFSAN/Office of Food Additive Safety, Summary of All GRAS Notices, available at <http://www.cfsan.fda.gov/~rdb/opa-gras.html>.

¹⁰ 21 U.S.C. 201, 348, 379e, and 601.

¹¹ 21 U.S.C. 342(a),(b)(3)-(4), and 601(m).

¹² See Meat Ingredients MOU, *supra* note 8.

Section 173.350 of FDA regulations specifies the conditions in which carbon monoxide can be safely used to displace oxygen in food and beverage packaging. This regulation authorizes the use of carbon monoxide for all food and beverage products at levels up to 4.5 percent,¹³ including in meat products, with the sole exception that carbon monoxide is categorically prohibited for such use in “fresh meat products.”¹⁴ It is well established that the specification prohibiting carbon monoxide in “fresh meat” is required under the FDCA because of the serious public health risks attributable to the capacity of carbon monoxide to mask spoilage and promote consumer deception under these conditions.

These public health risks and consumer deception implications further mandate label declaration of the use of carbon monoxide in fresh meat packaging. Although there are no grounds upon which FDA could lawfully allow this use of carbon monoxide, even assuming *arguendo* that FDA had such authority, the agency would be required to implement FDCA labeling provisions mandating that the presence and purpose of the carbon monoxide in the packaging system be disclosed.

Because the use of carbon monoxide to displace oxygen in packaging for fresh meat products violates a catalog of provisions of the FDCA and runs afoul of the agency’s own regulations, FDA’s failure to object to the Pactiv and Precept GRAS notifications constitutes unlawful agency action under the Administrative Procedure Act (“APA”).¹⁵ FDA’s Agency Response Letters are tantamount to unlawful color additive approvals, for they allow the use of deceptive colorants in violation of the FDCA and in the absence of a required color additive regulation.¹⁶ The agency’s failure to follow the statutorily-mandated procedures for color additive approval is an abuse of discretion, for as the Supreme Court has explained, “[i]t is rudimentary administrative law that discretion as to the substance of the ultimate decision does not confer discretion to ignore the required procedures of decisionmaking.”¹⁷

Moreover, FDA’s improper responses expressly allow a use of carbon monoxide that is explicitly prohibited by the agency’s own food additive regulation at section 173.350, in violation of the well-settled rule that an agency must follow its own regulations.¹⁸ As FDA has provided no justification for its deviation from that section’s prohibition against the use of carbon monoxide-containing packaging gases in fresh meat, its Agency Response letters represent arbitrary and capricious agency action. Treating similar situations differently is the

¹³ 21 C.F.R. 173.350(b)(1).

¹⁴ 21 C.F.R. 173.350(c).

¹⁵ See 5 U.S.C. § 706(2).

¹⁶ See 5 U.S.C. § 706(2)(C) & (D) (empowering courts to find unlawful any agency actions in excess of statutory limitations or without observance of procedures required by law).

¹⁷ *Bennett v. Spear*, 520 U.S. 154, 172 (1997).

¹⁸ See, e.g., *Mine Reclamation Corp. v. FERC*, 30 F.3d 1519, 1524 (D.C. Cir. 1994) (characterizing the “well-settled rule that an agency’s failure to follow its own regulations is fatal to the deviant action”).

essence of arbitrary and capricious agency action. The Court of Appeals for the District of Columbia Circuit has made clear that “[a]n agency must treat similar cases in a similar manner unless it can provide a legitimate reason for failing to do so.”¹⁹ For these reasons, FDA’s failure to object to the Pactiv and Precept GRAS notifications is unlawful under the APA.²⁰

In view of the serious public health issues presented and the requirements of the FDCA and APA, FDA has no legal authority to permit the use of carbon monoxide in fresh meat packaging, and the agency’s unlawful responses to the Pactiv and Precept GRAS notifications must be terminated immediately.

3. Applicable Legal Standards

a. Regulatory Framework Governing the Ingredients of Fresh Meat Products

Under a Memorandum of Understanding between FDA and FSIS implemented in January, 2000 (“Meat Ingredients MOU”), the two agencies adopted joint procedures permitting the expedited approval of meat product ingredients, including color additives, food additives, and GRAS substances.²¹ The new policy supplanted the longstanding procedures requiring independent and sequential premarket clearance first, by FDA, under the requirements of the FDCA, and second, by FSIS, under the requirements of the FMIA.

Under the FDCA, FDA has authority for making safety determinations with respect to food ingredients constituting “color additives,” “food additives,” and substances that are “generally recognized as safe” (“GRAS”), including those intended for use in fresh meat. Under the FMIA, FSIS has authority for making “suitability determinations” concerning ingredients intended for use in meat products.²² The FSIS “suitability” evaluation considers consumer protection issues specific to meat products, and may impose limitations on ingredient uses in meat that are not required for more general use in food. FSIS guidance provides that, “suitability relates to the effectiveness of the additive in performing the intended technical purpose of use, at the lowest level necessary, and the assurance that the conditions of use will not

¹⁹ *Independent Petroleum Ass’n v. Babbitt*, 92 F.3d 1248, 1258 (D.C. Cir. 1996). See also *Kent County, Delaware v. EPA*, 963 F.2d 391 (D.C. Cir. 1992); *Green Country Mobilephone, Inc. v. FCC*, 765 F.2d 235, 237 (D.C. Cir. 1985) (same).

²⁰ Similarly, FSIS’s failure to object to carbon monoxide as unsuitable for the purposes proposed in the Pactiv and Precept GRAS notifications contravenes the FMIA, its implementing regulations, and established USDA policy, and is likewise unlawful agency action under the APA.

²¹ Meat Ingredients MOU, *supra* note 8.

²² See 65 Fed. Reg. 3330 and Meat Ingredients MOU, *supra* note 8.

result in an adulterated product or one that misleads consumers."²³ Meat products may include only those ingredients that FSIS has expressly authorized.²⁴

Under well established FSIS policy, ingredients that function in fresh meat to conceal damage or inferiority, or give the appearance the product is better or of greater value than is the case are prohibited.²⁵ Consistent with this policy, FSIS not only has declined to authorize the use of color additives in fresh meat,²⁶ but also has issued rules explicitly prohibiting such use. For example, despite FDA's determination that "paprika" is safe, including for color additive purposes in food generally, FSIS has prohibited the use of paprika in fresh meat products.²⁷ FSIS justified the restriction on paprika as "necessary to assure that federally inspected meats and meat food products are not adulterated through the use of substances that conceal damage or inferiority or make the product appear to be better or of greater value than they are."²⁸

Under FDCA requirements, food ingredients that constitute either "food additives" or "color additives" are prohibited, including in fresh meat products, except where FDA has determined the ingredient to be safe under the conditions of intended use and has promulgated regulations authorizing such use.²⁹ Food ingredients that are established to be

²³ See Guidance on the Procedures for Joint Food Safety and Inspection Service (FSIS) and Food and Drug Administration (FDA) Approval of Ingredients and Sources of Radiation Used in the Production of Meat and Poultry Products," available at <http://www.fsis.usda.gov/OPPDE/rdad/FRPubs/00-022N/ApprovalofIngredients.htm>.

²⁴ 9 C.F.R. 424.21.

²⁵ See, e.g., 21 U.S.C. 601(m); 9 C.F.R. 424.23.

²⁶ FSIS regulations prohibit the use of color-imparting substances in meat products in the absence of authorizing regulations. See 9 C.F.R. 424.21(b)(3) ("No food ingredient, the intended use of which is to impart color in any meat or poultry product, shall be used unless such use is approved in 21 CFR Chapter I as a color additive . . . or in a regulation in this chapter."). While the FSIS regulation at 9 C.F.R. 424.22(a)(3) states that coloring matter and dyes other than those specified by regulation may be used if approved by the Administrator in specific cases, this approval process is available only for additives applied to meat mixed with rendered fat and to casings; this procedure is not a vehicle for approval of colorants to be used in fresh meat.

²⁷ 21 C.F.R. 73.340 and 73.345 (listing paprika and paprika oleoresin for use in coloring foods generally); 21 C.F.R. 182.10 (authorizing use of paprika for spice or other natural seasoning and flavoring purposes); 9 C.F.R. 424.23(a)& (b).

²⁸ 34 Fed. Reg. 20386 (December 31, 1969) (Final Rule); see also 65 Fed. Reg. 51758, 51759 (August 25, 2000) (FDA recognizing the established policy prohibiting the use of paprika in meat on consumer protection and public health grounds).

²⁹ 21 U.S.C. 348 (requiring FDA premarket approval of food additives that are not food contact substances, and authorizing such approval only where there is reasonable certainty that the substance is not harmful under the intended conditions of use); 21 U.S.C. 379e (requiring FDA premarket approval and listing of color additives, and authorizing such listing only where the substance is suitable and safe under the conditions of intended use).

GRAS under the conditions of intended use are excluded from the FDCA premarket clearance requirements that apply to “food additives” but not from those that apply to “color additives.” This means that, for a food ingredient that is established to be GRAS under certain conditions of use, the food ingredient may lawfully be used under such conditions without an authorizing food additive regulation. In contrast, for the same ingredient to be used for color additive purposes, FDA must promulgate regulations listing the food ingredient for specified conditions of color additive use. For example, while the established GRAS status of paprika for seasoning purposes eliminates the need for a food additive regulation to authorize seasoning uses, paprika could not be used under similar conditions for coloring purposes in the absence of the FDA regulations listing paprika specifically for color additive purposes.³⁰

The Meat Ingredients MOU implements streamlined premarket clearance procedures, but reflects no change in the legal standards governing authorizing the use of food additives, color additives, or GRAS substances under the FDCA and FMIA.³¹ Under the new coordinated FDA/FSIS procedures for expedited food ingredient review, petitions for food additives and color additives must be submitted to FDA, which is responsible for promulgating regulations authorizing these substances when they are safe under the intended conditions of use. Where the intended conditions of use encompass fresh meat products, the MOU provides that FDA and FSIS will jointly review petitions, and final FDA regulations will specify appropriate restrictions concerning such uses, as recommended by FSIS.³²

The Meat Ingredients MOU establishes fast track procedures for agency review of GRAS notifications for non-color additive uses in meat products. The coordinated FDA/FSIS procedures provide that GRAS notifications that are submitted to FDA be reviewed concurrently by FSIS for purposes of making suitability determinations. The MOU provides that the FDA letter responding to a GRAS notifier may convey FSIS concerns about the suitability of the ingredient use in meat products, and may specify restrictions on use that have been recommended by FSIS.³³ Color additives cannot be reviewed under these coordinated procedures for GRAS notifications. Under the FDCA, FDA can authorize color additives only under conditions that have been determined to be safe and are specified in regulations issued

³⁰ 21 C.F.R. 73.340 (listing paprika for “the coloring of foods generally, in amounts consistent with good manufacturing practice . . .”); see also 21 C.F.R. 73.345 (listing paprika oleoresin for color additive purposes).

³¹ Meat Ingredients MOU at 4 (stating that “[t]he provisions of this MOU are not intended to add to or detract from any of the authorities provided to either FDA or FSIS by the [FDCA or FMIA] . . . or the regulations promulgated by each agency under such authorities” and “[e]ach agency reserves the authority to review, independently of the other, matters of concern to their respective authorities.”).

³² Meat Ingredients MOU at 4.

³³ Meat Ingredients MOU at 5.

through notice and comment rulemaking procedures.³⁴ FSIS lacks authority to authorize the use of any color additive that has not been approved by FDA through this procedure.³⁵

4. The FDCA Prohibits the Use of Carbon Monoxide in Fresh Meat Packaging

a. Carbon Monoxide Constitutes an Unapproved Color Additive

Under FDCA section 721, adopted under the Color Additive Amendments of 1960, color additives are prohibited from use in food except under the defined conditions of use specified in by FDA regulations "listing" the particular color additive.³⁶ Currently, there are no FDA regulations authorizing the use of carbon monoxide in fresh meat, as required by FDCA section 721.

Section 201(t)(1) of the FDCA defines "color additive" to mean any "substance made by a process of synthesis . . . or otherwise derived, with or without intermediate or final change of identity, . . . and when added or applied to a food . . . or to the human body . . . is capable (alone or through reaction with other substance) of imparting color thereto . . ."³⁷

Under well established FDA policy, "color additives" include substances that impart color through chemical reactions occurring after the substance is applied under the intended conditions of use. FDA has explained that "any chemical that reacts with another substance and causes formation of a color may be a color additive."³⁸ For example, FDA has regulated colorless ingredients of sunless tanning lotions and hair dyes as color additives where these substances participate in color imparting reactions with chemicals naturally present in skin and hair during application.³⁹

³⁴ 21 U.S.C. 379e.

³⁵ Under the Meat Ingredients MOU, FSIS lacks authority even to independently authorize the use of food ingredients in meat products that are currently approved under FDA regulations, where the conditions of use do not expressly encompass meat and poultry products. The MOU specifies that where the regulation does not specifically authorize uses in meat and poultry products, FSIS first would be required to obtain a written statement from FDA confirming the scope of the agency's earlier safety determination and expressing no objections with respect to the safety of the proposed conditions of use in meat products.

³⁶ 21 U.S.C. 371e.

³⁷ 21 U.S.C. 321(t)(1).

³⁸ See, e.g., "Color Additives: FDA's Regulatory Process and Historical Perspectives," reprinted from *Food Safety Magazine* (October/November 2003) ("Color Additives"), available at <http://www.cfsan.fda.gov/~dms/col-regu.html>.

³⁹ See, e.g., 21 C.F.R. 73.2150 (regulating dihydroxyacetone ("DHA") as color additive where the colorless substance, when applied to the skin, reacts with natural skin proteins resulting in the formation of a brown coloring on the skin surface); 21 C.F.R. 73.2396 (regulating lead acetate (continued...))

FDA has also has regulated ingredients of food as color additives when the ingredient subsequently participates in color-imparting chemical reactions under the conditions of intended use. For example, ingredients of animal feed intended for consumption by poultry and salmon have been regulated as color additives where the ingredients participate in metabolic reactions which intensify the color of the animal tissues intended for use as human food (e.g., intensified gold in egg yolks and red in salmon fillets).⁴⁰

FDA has recognized that ingredients which impart color to meat products through chemical reactions with the naturally occurring myoglobin in meat tissues are appropriately regarded as "color additives" within the meaning of FDCA section 201(t)(1). Specifically, in responding to a citizen petition requesting FDA to regulate nitrites in cured meat under FDCA section 721, FDA evaluated the color-imparting effects of nitrite under the "color additive" definition of the Act. While concluding that a "prior sanction" authorizing the use of nitrite in cured meat ultimately nullified the requirements of FDCA section 721 in this context,⁴¹ FDA determined that nitrites did, in fact, "impart color" within the meaning of the color additive definition, as a result of reactions occurring with myoglobin. FDA stated, "nitrites 'impart' color . . . by reacting with a substance naturally present in the meat to form a third substance that gives the meat a reddish appearance . . . The fact that the color given meat by nitrites is similar to the natural color of meat does not warrant the conclusion that the effect of nitrites is merely to 'fix,' rather than 'impart,' color."⁴²

for use in hair dye as color additive); 21 C.F.R. 73.2110 (regulating bismuth citrate as color additive for use in hair dye); see also "Color Additives," *supra* note 39.

⁴⁰ See, e.g., 21 C.F.R. 73.275 (regulating dried algae meal in chicken feed as color additive to enhance the yellow color of chicken skin and egg yolks); 21 C.F.R. 73.295 (regulating tagetes/Aztec marigold meal and extract in chicken feed as color additive to enhance the yellow color of chicken skin and egg yolks); 21 C.F.R. 73.35 (regulating astaxanthin meal in salmon feed as a color additive to enhance the pink to orange-red color of the fish flesh); 21 C.F.R. 73.185 (regulating haematococcus algae meal in salmon feed as a color additive to enhance the pink to orange-red color of the fish flesh).

⁴¹ FDA ultimately concluded that the existence of a prior sanction for nitrites established under FDCA section 201(s)(4) provided an adequate legal basis for maintaining the established nitrite policy. The agency concluded that the long history of safe use of nitrites, the enhanced food safety of cured meat products, and consumer familiarity with the distinctive coloration of cured meats justified its decision to uphold the nitrite prior sanction. 45 Fed. Reg. 77043, 77045 (November 21, 1980) (Withdrawal of Proposed Rule). In contrast to nitrites, not only has no prior sanction been established for carbon monoxide in fresh meat, but such use is explicitly prohibited under section 173.350 of FDA regulations. In addition, carbon monoxide is not used to cure meat or otherwise preserve the safety and quality of meat. To the contrary, carbon monoxide obscures the natural coloration of meat and gives the appearance of freshness and safety when the natural colors would indicate otherwise.

⁴² Letter from Donald Kennedy, Commissioner of Food and Drugs, to William B. Schultz, Public Citizen Litigation Group, at 12 (June 29, 1979) (Attachment 2).

Specifically, FDA determined that, in curing meats, nitrites function to displace water molecules that bind naturally to myoglobin, forming nitric oxide myoglobin, which imparts a red color to the meat. In contrast, in fresh meat, myoglobin naturally binds with oxygen to form oxymyoglobin under ambient conditions. In addition, when cured meat is cooked, nitric oxide myoglobin yields nitrosyl hemochrome, which is pink in color. In contrast, when fresh meat is cooked, oxymyoglobin yields denatured metmyoglobin, which is brown in color. FDA characterized the color imparting effects of nitrites in the context of cooked meat as follows, “[w]ere it not for the use of nitrites, the meat would have a brown color after heating rather than the pink attributed by the presence of nitrosyl hemochrome. Nitrites thus ‘impart’ color by giving the meat a color after heating that it would not otherwise have.”⁴³

Like nitrites, carbon monoxide in fresh meat packaging imparts color to meat through chemical reactions with the myoglobin naturally occurring in meat tissues. Myoglobin, which occurs in the muscle fibers of living animals, is a biological oxygen carrier like hemoglobin, to which it is chemically related.⁴⁴ Like the hemoglobin in circulating blood, myoglobin functions to deliver oxygen to the tissues of living animals.⁴⁵ Just as the redness of blood varies with the degree to which hemoglobin is oxygenated, so also does the redness of meat vary with the oxygenation of myoglobin. As the myoglobin in fresh cut meat binds naturally with oxygen under ambient conditions, oxymyoglobin is formed, and is responsible for the red color indicative of fresh meat. Over time, the oxymyoglobin participates in further reactions with oxygen, gradually oxidizing to form metmyoglobin, which is browner in color. As oxidation advances, the freshness and safety of fresh meat decreases in relationship to the progressive browning of meat color. Eventually, meat takes on the browned color that consumers have long relied upon to indicate that meat is spoiled and unsafe to consume.

When the oxygen in fresh meat packaging is displaced by carbon monoxide, the natural coloration provided by meat pigments is masked. Carbon monoxide binds firmly to myoglobin sites that otherwise would be bound more gently by oxygen, forming carboxymyoglobin in place of oxymyoglobin. Carboxymyoglobin imparts an intense red color to the meat which, in contrast to oxymyoglobin, resists the further reactions with oxygen that would form metmyoglobin. In this regard, carbon monoxide is categorically different from antioxidant color preservatives, which simply inhibit the oxygenation of myoglobin in meat, rather than reacting with the myoglobin to form a new chemical substance.

Just as breathing carbon monoxide endangers living animals through its stubborn displacement of oxygen in circulating hemoglobin, adding carbon monoxide to fresh meat endangers consumers by stubbornly displacing oxygen in meat myoglobin. Carboxymyoglobin imparts a sustained bright red color to meat that simulates the appearance of freshness and safety in meat when the natural pigments would warn consumers otherwise.

⁴³ 44 Fed. Reg. 75659, 75660 (December 21, 1979) (Proposed Rule).

⁴⁴ Encyclopedia of Chemical Technology, Vol. 14, at 895 (4th Ed. 1995).

⁴⁵ *Id.*, Vol. 16, at 765.

b. The Pactiv and Precept GRAS Notifications Cannot Support Fast Track Listing of Carbon Monoxide for Color Additive Purposes

While the proviso at section 721(b)(4) provides for fast-track FDA approval for color additives where FDA previously has determined the ingredient to be GRAS, the abbreviated procedures have no application in the context of carbon monoxide use in fresh meat packaging. Section 721(b)(4) provides that, "a color additive shall be deemed to be suitable and safe for the purpose of listing under this subsection for use generally in or on food, while there is in effect a published finding of the secretary declaring such substance exempt from the term 'food additive' because of its being generally recognized by qualified experts as safe for its intended use, as provided in section 201(s)." This proviso was adopted to ensure that redundant regulatory approval procedures would not be compelled by the Color Additive Amendments made to the FDCA in 1960 for such common household food ingredients as salt, vinegar, and natural spices, which FDA had determined were GRAS.⁴⁶ Indeed, in FDA's responses to other GRAS notifications for substances whose use may constitute that of a color additive in certain applications, the agency makes clear that although some uses may be GRAS, other uses of the same substance will require premarket review, approval, and listing as a color additive.⁴⁷

Section 721(b)(4) never was intended to provide fast track approval for such substances as carbon monoxide, which have historically been banned for use in fresh meat, much less lacking any history of safe use in such food. In addition, even if the unlawful Pactiv and Precept GRAS notifications were valid, they would provide no lawful basis for fast track listing of carbon monoxide for color additive uses in fresh meat products. FDA has repeatedly emphasized that the agency's "no objection" letter responding to a GRAS notification does not constitute an FDA "published finding" that an ingredient is GRAS, for purposes of FDCA section 721(b)(4).⁴⁸ Moreover, even where FDA has, in fact, issued a "published finding" of GRAS status, the provision makes no change in the standards for safety and suitability that must be satisfied for a color additive to be approved by FDA. Neither section 721 nor any other FDCA provision authorizes FDA to list a color additive that is unsafe or promotes consumer deception under the conditions of intended use.

In sum, the FDA lacks the legal authority to condone the GRAS status of carbon monoxide uses in fresh meat packaging. The FDCA obligates FDA to withdraw its responses to the GRAS notifications submitted by Pactiv and Precept and prohibit all such use of carbon

⁴⁶ See H.R. Report No. 86-1761, at 15 (1960) ("House Report").

⁴⁷ See, e.g., letter from Laura M. Tarantino, Acting Director, CFSAN Office of Food Additive Safety, to George A. Burdock, Ph.D., Burdock Group (February 7, 2005), at 4, available at <http://www.cfsan.fda.gov/~rdb/opa-g156.html> (Agency Response Letter to GRAS notice for tomato lycopene extract).

⁴⁸ See, e.g., letter from Laura M. Tarantino, Acting Director, CFSAN Office of Food Additive Safety, to Dr. Dore, Cyanotech Corp. (Oct. 6, 2003), (Agency Response Letter to GRAS Notice No. GRN 000127), n.2, available at <http://www.cfsan.fda.gov/~rdb/opa-g127.html> ("no questions" response "does not constitute a 'finding of the Secretary' within the meaning of section 721(b)(4) of the [FDCA]").

monoxide in the absence of authorizing color additive regulations. Even if such color additive petitions were submitted, however, FDA would be unauthorized to list carbon monoxide as a color additive for use in fresh meat. Carbon monoxide fails to meet the statutory criteria of safety and suitability, as established for color additives under FDCA section 721.

c. The Use of Carbon Monoxide in Fresh Meat Packaging Cannot Satisfy the Safety and Suitability Requirements for Color Additive Listing

Section 721(b)(1) of the FDCA authorizes FDA to promulgate a regulation listing a color additive for use in food only “if and to the extent that such additives are suitable and safe for any such use when employed in accordance with such regulations.”⁴⁹ Further, section 721(b)(6) prohibits FDA from listing a color additive for a proposed use if that use “would promote deception of the consumer in violation of this Act or would otherwise result in misbranding or adulteration within the meaning of this Act.”⁵⁰ These provisions operate both independently and in conjunction to prohibit the listing of carbon monoxide for use in fresh meat packaging, for this use is neither safe nor suitable precisely because it promotes deception that results in serious food safety concerns.

i Colorants for Meat Have Never Been Approved by FDA or FSIS, Because They Would Promote Deception by Making Meat Appear Fresher Than It Is

Ensuring prevention of deception was an overarching principle behind the Color Additive Amendments, as revealed in the text and legislative history of those amendments, FDA implementing regulations, and interlocking FSIS meat additive regulations and suitability determinations. Significantly, Congress and FDA’s predecessor agency were particularly concerned about the use of deceptive colorants in meat. “Examples of coloring practices that would promote deception of the consumer in violation of the basic act were cited by the Secretary of Health, Education, and Welfare as follows: . . . (4) the use of artificial color in stale red meat to make it appear fresh.”⁵¹ Additionally, Section 204 of those Amendments mandates that “[n]othing in this Act shall be construed to exempt any meat or meat food product, poultry or poultry product, or any person from any requirement imposed by or pursuant to the Meat Inspection Act of March 4, 1907, 34 Stat. 1260, as amended or extended . . .”⁵² Thus, although the use of carbon monoxide in fresh meat packaging is relatively new, it gives rise to the precise type of deception anticipated and opposed by the drafters of the Color Additive Amendments.

⁴⁹ 21 U.S.C. 379e(b)(1).

⁵⁰ 21 U.S.C. 379e(b)(6).

⁵¹ House Report at 17.

⁵² Pub. Law 86-618, 71 Stat. 441.

No coloring agents are authorized for use in fresh meats in FSIS's regulations enumerating substances permitted for use in meat and poultry products.⁵³ To the contrary, additives that have been recognized to impart color to fresh meat have been affirmatively prohibited.⁵⁴ The prohibition against colorants in fresh meat dates back to even before the enactment of the Color Additive Amendments of 1960. Before that time, colorants in meat were prohibited under the adulteration provisions of the FDCA and FMIA,⁵⁵ upon which the antideception provisions of the Color Additive Amendments were derived⁵⁶ and which continue to function as an alternative statutory basis upon which colorants in fresh meat are prohibited. For example, the ban on the use of sodium sulfite in meat products⁵⁷ has been documented as early as 1943, when FDA explained that "[d]ue to the effect of sulfites on meat products, that is, old and dull colored meat can be rendered red and fresh looking, we are of the opinion that its use in meat is likely to render such meat adulterated under the provisions of the Food, Drug, and Cosmetic Act in that damage and inferiority are concealed or the product made to appear better or of greater value than it is."⁵⁸ FSIS has never wavered from this position, and in its press release regarding a 1998 criminal action securing felony sentences for violators who used sodium sulfite, FSIS emphasized that "[s]odium sulfite is banned as a preservative in meat and poultry products because it masks the spoilage and color change due to aging."⁵⁹ FSIS banned the use of paprika for this same reason,⁶⁰ explaining that the spice "preserv[es] the red color

⁵³ 9 C.F.R. 424.21(c); *see also* FSIS Directive 7120.1.

⁵⁴ *See* 9 C.F.R. 424.23(a).

⁵⁵ 21 U.S.C. 342(b)(3)&(4), 601(m)(8) (providing, in relevant part, that a food is adulterated "if damage or inferiority has been concealed in any manner; or if any substance has been added thereto or mixed or packed therewith so as to . . . make it appear better or of greater value than it is.").

⁵⁶ *See* House Report at 16-17 (explaining that, with respect to Section 721(b)(6), "[i]t should be emphasized that we are dealing here solely with deception which would violate the law," and citing sections 402(b)(3) & (4) of the FDCA as the relevant statutory provisions implicated by 721(b)(6)).

⁵⁷ 9 C.F.R. 424.23(a)(3) (prohibiting the use of sulfurous acid and salts of sulfurous acid in or on any meat because they conceal damage or inferiority or make products appear of better or of greater value than they are).

⁵⁸ *See* letter from Joseph Callaway, Jr., Acting Chief, Division of State Cooperation, to Wayne B. Adams, Acting State Food and Drug Commissioner, Nevada, October 14, 1943, at 3 (Attachment 3). Notably, that letter acknowledged that a number of state laws prohibited the use of sulfites in sausage at that time, even where the additive was allowed in other foods, because "it has been generally held that the use of sulfites in meat and meat products violates a provision in most food laws against the use of any substance to conceal damage or inferiority or cause the product to appear of better or greater value than it is." *Id.* at 1.

⁵⁹ *See* <http://www.fsis.usda.gov/oa/news/1998/cr98-10.htm>.

⁶⁰ 9 C.F.R. 424.23(a). That rule also prohibits the use of sorbates because their use "conceals damage and inferiority, i.e., the fact that the products are decaying because of bacterial action, and makes the products appear better and of greater value than they are in view of their (continued...)

characteristic of fresh meat even after the articles have begun to spoil, and thereby conceals damage or inferiority and makes them appear to be better and of greater value than they are.”⁶¹

There is no conceivable distinction between the effect of sodium sulfite or paprika and that of carbon monoxide on fresh meat. To allow carbon monoxide in fresh meat packaging would constitute an unjustifiable departure from prior regulatory action on additives serving a virtually identical function. Carbon monoxide has similarly been shown to mask spoilage and color change due to aging by imparting an artificial red color that mimics that of fresh meat. The chemical thereby conceals damage and inferiority and makes meat appear to be of greater value than it is, within the meaning of the adulteration provisions of the FDCA and FMIA. As such, carbon monoxide in fresh meat packaging would promote consumer deception. Accordingly, FDA is prohibited by section 721(b)(6) of the FDCA from listing carbon monoxide for use in fresh meat packaging as a color additive.

This is precisely the conclusion reached by FSIS during the course of its review of the Precept GRAS notification. In a letter from the director of FSIS’s Labeling and Consumer Protection Staff to FDA’s Office of Food Additive Safety, FSIS explained:

The Precept Foods MAP system stabilizes the color of the meat and, therefore, by affecting one of the sensory properties (i.e., appearance) used in assessing the quality of a meat product has the potential to mislead consumers into believing that the product they are purchasing is fresher than it actually is.

...

In summary, it is our opinion that the use of the Precept Foods MAP system described in GRAS Notice No. GRN 000143 for use with case-ready fresh cuts of meat and ground meat could potentially mislead consumers into believing that they are purchasing a product that is fresher or of greater value than it actually is and may increase the potential for masking spoilage.⁶²

The FDA public record produced in response to a Freedom of Information Act (“FOIA”) request is devoid of an explanation of why its Agency Response Letter to the Precept GRAS notification, issued only three months after FSIS expressed the conclusions above, states that FSIS concluded that the Precept MAP system is acceptable for packaging fresh meat.⁶³ In any event, whatever

decomposing condition.” 35 Fed. Reg. 15552, 15553 (October 3, 1970) (Revision Pursuant to Wholesome Meat Act).

⁶¹ 33 Fed. Reg. 15027 (October 8, 1968) (Proposed Rule).

⁶² Letter from Robert C. Post, Ph.D., Director, Labeling and Consumer Protection Staff, to Dr. Lane Highbarger, Office of Food Additive Safety, CFSAN, FDA, April 28, 2004 (Attachment 4).

⁶³ Agency Response Letter to GRAS Notice No. GRN 000143, at 3.

transpired during that narrow time period cannot justify a determination that the use of carbon monoxide is generally recognized as safe, as a matter of law.

ii. FDA Has Failed to Demonstrate that Carbon Monoxide in Fresh Meat Packaging Would Be Safe Under Actual Conditions of Use

A central intent of Congress in enacting the Color Additive Amendments was to ensure that such additives will be safe under actual conditions of use. The legislative history emphasizes the overarching “safe for use” principle, which is the “scientifically sound principle that we must consider conditions of use when passing on suitability and safety of a color additive.”⁶⁴ FDA is required to consider actual conditions of consumer use when evaluating a color additive, and must have concrete evidence that the additive will be used safely. The House Report explains that a color additive may be listed for use only when it is *shown* that it may be safely used under the conditions prescribed by regulation.⁶⁵ Moreover, the regulatory definition of “safe” with respect to color additives “means that there is convincing evidence that establishes with reasonable certainty that no harm will result from the intended use of the color additive.”⁶⁶

Neither FDA nor FSIS have evidence establishing that carbon monoxide in fresh meat packaging is safe under the actual conditions of use. To the contrary, the evidence demonstrates that the use of carbon monoxide in anaerobic packaging systems for fresh meat poses genuine food safety risks under real-world conditions. Significantly, even FDA itself has emphasized the substantial food safety concerns that accompany foods – particularly meats – packaged with oxygen-displacing gases, such the carbon monoxide-containing modified atmospheres that are the subjects of GRN 83 and 143.⁶⁷

FDA has devoted a portion of its Food Code to the subject of reduced oxygen packaging (“ROP”). The agency explains that an “anaerobic environment, usually created by ROP, provides the potential for growth of several important pathogens.”⁶⁸ Specifically, “[i]f products in ROP are subjected to mild temperature abuse, i.e., 5°-12°C (41°-53°F), at any stage during storage or distribution, foodborne pathogens, including *Bacillus cereus*, *Salmonella* spp., *Staphylococcus aureus*, and *Vibrio parahaemolyticus* can grow slowly. Marginal refrigeration that does not facilitate growth may still allow *Salmonella* spp., *Campylobacter* spp., and *Brucella* spp. to survive for long periods of time.”⁶⁹

⁶⁴ S. Report No. 86-795, at 4 (1959).

⁶⁵ House Report at 25 (emphasis added).

⁶⁶ 21 C.F.R. 70.3(i).

⁶⁷ Food and Drug Administration, Food Code 544 (2005) (“ROP [reduced oxygen packaging] which provides an environment that contains little or no oxygen . . . raises many microbiological concerns.”).

⁶⁸ *Id.* at 546.

⁶⁹ *Id.* at 547.

Of particular concern in ROP are *Clostridium botulinum* and *Listeria monocytogenes*. FDA emphasizes,

If present, *C. botulinum* could potentially grow and render toxigenic a food packaged and held in ROP because most other competing organisms are inhibited by ROP. Therefore, the food could be toxic yet appear organoleptically acceptable. This is particularly true of psychrotrophic strains of *C. botulinum* that do not produce tell-tale proteolytic enzymes. Because botulism is potentially deadly, foods held in anaerobic conditions merit regulatory concern and vigilance.⁷⁰

Despite the agency's cautionary language in the Food Code, FDA has failed to exhibit appropriate regulatory concern and vigilance in failing to object to the proposed use of carbon monoxide in anaerobic packaging for fresh meat. No material distinction exists between fresh meats packaged in ROP at retail and fresh meat packaged pursuant to GRN 83 and GRN 143 such that FDA could reasonably ignore the safety concerns it stresses in the Food Code. Yet, there is no indication that the agency considered imposing, as a condition for safe use of carbon monoxide in anaerobic fresh meat packaging, any of the safety barriers it emphasized in the Food Code for ROP-packaged products, including meat. Most notably, FDA repeatedly expresses the need for temperature control where ROP-packaged products such as fresh meat are not treated to prevent microbial contamination.⁷¹ The agency would mandate that all foods in ROP which rely on refrigeration as a barrier to microbial growth bear the statement, on the principal display panel in bold type on a contrasting background, "Important – Must be kept refrigerated at 5°C (41°F)."⁷² Inexplicably, however, FDA imposed no such refrigeration statement as a condition of safe use of carbon monoxide in anaerobic packaging for fresh meat.

As a practical matter, however, such a refrigeration advisory would have little effect, given fact that temperature abuse, both during distribution and consumer handling of fresh meat, and related food safety concerns are well documented.⁷³ Such abuse is compounded for

⁷⁰ *Id.* at 548.

⁷¹ *Id.* ("Processors of products using ROP should be cautious if they plan to rely on refrigeration as the sole barrier that ensures product safety. This approach requires very rigorous temperature controls and monitored refrigeration equipment. If extended shelf-life is sought, a temperature of 3.3°C (38°F) or lower must be maintained at all times to prevent the outgrowth of *C. botulinum* and the subsequent production of toxin.").

⁷² *Id.* at 551.

⁷³ See, e.g., Labuza, T.P. and Fu, B., "Use of Time/Temperature Integrators, Predictive Microbiology, and Related Technologies for Assessing the Extent and Impact of Temperature Abuse on Meat and Poultry Products, 15 J. Food Safety 201-227 (1995) (Attachment 5), at 202 ("Unfortunately, the existing distribution channel is not well equipped for the optimum control of temperature during the distribution and display of refrigerated foods. Temperature abuse is common throughout the distribution and retail markets, with the temperature in 21% of household refrigerators often higher than 10°C. Recent data suggested that 33% of retail (continued...)

meats packaged in modified atmospheres with an intended longer shelf life, which provides more opportunities for the food to encounter abusive temperature variation during distribution and storage, thereby increasing the likelihood of microbial spoilage.⁷⁴ FDA acknowledges that “[t]emperature abuse is common throughout distribution and retail markets.”⁷⁵ The agency cites published surveys indicating that refrigeration practices at retail need improvement, and cautioned that “[i]t must be assumed . . . for purposes of assessing risk, that occasionally temperatures of 10°C (50°F) or higher may occur for extended periods” in warehouses and transport vehicles in U.S. distribution chains.⁷⁶

Of even greater concern are consumer handling practices. FDA observes that “[c]onsumers often cannot, or do not, maintain adequate refrigeration of potentially hazardous foods at home Under the best of circumstances, home refrigerators can be expected to range between 5° and 10°C (41°-50°F).”⁷⁷ Thus, while the need for strict temperature control has been emphasized where fresh meat is packaged with oxygen-displacing gases,⁷⁸ including

refrigerated foods were held in display cases above 7°C and 5% were held above 13°C. Temperatures were even higher in southern market regions. Serious microbial stability problems exist because of the frequency of temperature abuse.”) (citations omitted).

⁷⁴ See, e.g., Farber, J.M., “Microbiological Aspects of Modified-Atmosphere Packaging Technology – A Review,” 54 J. Food Protection 58-70 (January 1991) (Attachment 6), at 58 (stating that microbiological safety issues have been raised about modified-atmosphere packaged foods mainly because of “the fact that the extended shelf life of many MAP products may allow extra time for . . . pathogens to reach dangerously high levels in a food”).

⁷⁵ FDA Food Code, *supra* note 68, at 547.

⁷⁶ *Id.*; see also Greer, G.G., et al., “Evaluation of the Bacteriological Consequences of the Temperature Regimes Experienced by Fresh Chilled Meat During Retail Display,” 27 Food Research Int’l 371-377 (1994) (Attachment 7) (reporting survey of commercial retail cases finding that recommended temperatures of 4°C or below cannot be maintained throughout existing retail cabinets).

⁷⁷ FDA Food Code, *supra* note 68, at 550.

⁷⁸ See, e.g., Lambert, A.D., et al., “Shelf Life Extension and Microbiological Safety of Fresh Meat – A Review,” 8 Food Microbiology 267-297 (1991) (Attachment 8), at 272 (the data “emphasizes [sic] the need for strict temperature-control of meat packaged under modified atmospheres” because such packaging “favors the growth of potential pathogenic clostridia under temperature abuse conditions”); see also Farber, *supra* note 74, at Table 1 (listing among the potential disadvantages of MAP the fact that temperature control is necessary); Nissen, H., et al., “Comparison Between the Growth of *Yersinia enterocolitica*, *Listeria monocytogenes*, *Escheria coli* O157:H7 and *Salmonella* spp. in Ground Beef Packed by Three Commercially Used Packaging Techniques,” 59 Int’l. J. Food Microbiology 211-220 (2000) (Attachment 9), at 212 (finding that *Salmonella* strains in inoculated ground beef stored at 10°C for 5 and 7 days grew to a higher number in a high carbon dioxide/low carbon monoxide gas mixture than in a high oxygen mixture, and stating that in such systems, “[a]t abuse temperatures (>8°C) *Escherichia coli* O157:H7 and *Salmonella* spp. also may grow and increase the health risk to the consumers.”).

carbon monoxide, the reality is that the necessary temperature criteria to ensure safety cannot be satisfied under actual conditions of use.

Given FDA's extreme vigilance over anaerobic packaging in other contexts, it is perplexing that the agency failed to impose strict controls on the use of carbon monoxide and other gases to displace oxygen in fresh meat packaging. With regard to low-acid and acidified canned foods, where the anaerobic environment can allow *Clostridium botulinum* spores to flourish, FDA imposes extensive regulations articulating the equipment, controls, manufacturing, processing, and packing procedures which are required to ensure the production of a safe product.⁷⁹ Yet in response to carbon monoxide-containing anaerobic packaging for fresh meat, which is known to potentially host a wide range of pathogens, including *Clostridium botulinum*, FDA has imposed no production controls whatsoever, and has failed even to require labeling concerning the need for refrigeration.

It is difficult to conceive of how the controls necessary to ensure the safe use of oxygen-displacing carbon monoxide in fresh meat packaging could be established without the promulgation of a food additive regulation, which would provide clear safety criteria to meat packagers using the technology and which would also serve as a touchstone for enforcement efforts to monitor its safe use. Given the realities of temperature abuse in current meat distribution systems, however, even criteria documented by regulation could not be satisfied under actual conditions of use.

Nor are USDA's extensive HACCP criteria sufficient to assure the safe use of carbon monoxide to displace oxygen in fresh meat packaging. USDA explains that questions to be considered in a hazard analysis include the following: "Is it likely that the food will contain pathogens and are they likely to increase during the times and conditions under which the food is normally stored before being consumed?" and "Does the method of packaging affect the multiplication of pathogenic microorganisms and/or the formation of toxins?"⁸⁰ For fresh meats packaged with oxygen-displacing gases including carbon monoxide, the answer to both questions is a resounding "yes," suggesting a potential food hazard and revealing the anaerobic packaging step to be a critical control point ("CCP"). Yet no appropriate critical limits for preventive measures associated with this CCP appear to have been established, as is required under the HACCP rule. Significantly, the microbiological performance standards for raw products under the HACCP rule involve only *Escherichia coli* and *Salmonella*,⁸¹ which are both aerobic organisms, for the assumption underlying the establishment of those criteria appears to be that the meats will be packaged in oxygen-containing environments. Accordingly, there appear to be no process control verification criteria that test for the presence of the anaerobic pathogens of concern when meat is packaged without oxygen.

⁷⁹ See 21 C.F.R. 108, 113, 114.

⁸⁰ 61 Fed. Reg. 38806, 38815 (July 25, 1996) (HACCP Final Rule).

⁸¹ 9 C.F.R. § 310.25.

The serious food safety concerns about anaerobic packaging are substantially magnified when carbon monoxide is included among the oxygen-displacing gases. In such packaging systems, not only does the anaerobic environment inhibit aerobic spoilage organisms that provide the indications of spoilage to which consumers are accustomed,⁸² but the color-imparting effect of the carbon monoxide also masks the natural color change of meat due to aging and deceptively suggests freshness well past the microbial shelf life of the meat.

It has been extensively documented that appearance – most notably, meat color – is the primary consideration of consumers in selecting meat and judging freshness.⁸³ By imparting a color resembling that of fresh meat, carbon monoxide in meat packaging deprives

⁸² See Labuza and Fu, *supra* note 73, at 203 (“A major question for chilled and/or MAP meat and poultry products is whether organoleptic spoilage due to chemical or microbial action will occur before the pathogen numbers or toxin levels become a risk when a product undergoes cycling or abuse temperatures.”); see also Hintlian, C.B. and Hotchkiss, J.H., “The Safety of Modified Atmosphere Packaging: A Review – Do Modified Atmospheres Enhance Pathogenesis But Delay Signs of Spoilage?” 40 Food Technology 70-76 (December 1986) (Attachment 10), at 75 (“The presence of air in packaged foods supports the growth of aerobic spoilage organisms. . . . In refrigerated products, this noxious warning by spoilage organisms is a critical safety factor since it serves to alert the consumer of temperature abuse and to prevent the consumption of a product which may also contain pathogens. Because anoxic MAs can favor the growth of facultative anaerobes and/or obligate organisms, packaging of foods in oxygen-excluded MAs could result in the loss of this safety factor.”).

⁸³ See, e.g., American Meat Science Association Guidelines for Meat Color Evaluation, available at <http://www.meatscience.org/Pubs/factsheets/M9110228.pdf>, at 3 (“The color of muscle foods is critically appraised by consumers and often is their basis for product selection or rejection.”); National Pork Board/American Meat Science Association Facts: Modified Atmosphere Packaging (MAP): Microbial Control and Quality, available at http://www.porkscience.org/documents/Other/Q-MAP-MICROBIAL_CONTandQUAL.pdf, at 3 (“Meat color is the single greatest appearance factor that determines whether or not a meat cut will be purchased”) (citation omitted); Kohls, L.I., *et al.*, “A Comparison of Five Different Modified Atmosphere Package Methods for Retail Display-Ready Ground Beef,” 2001 Animal Sciences Research Report, Colorado State University, available at <http://ansci.colostate.edu/dp/msfs/lik011.pdf>, at 1 (“Consumers view color as one of the most important attributes of fresh beef when making a decision to purchase retail product. Color, therefore, determines appeal of the product in the retail case and consumer acceptability.”); Jeremiah, L.E., *et al.*, “Beef Color as Related to Consumer Acceptance and Palatability,” 37 Journal of Food Science 476-479 (1972) (Attachment 11), at 476 (“Consumer studies have shown that physical appearance of a retail cut in the display case is the most important factor determining retail selection of meat products. Consumers select meat cuts primarily for leanness and then for appearance and freshness, with judgments for the latter two attributes based primarily on brightness of color.”) (citations omitted); Liu, Q., *et al.*, “Titration of Fresh Meat Color Stability and Malondialdehyde Development with Holstein Steers Fed Vitamin E-Supplemented Diets,” J. Anim. Sci. 1996, 74:117-126 (Attachment 12), at 117 (“Meat color is the main factor affecting beef product acceptability at retail points of purchase.”) (citation omitted).

consumers of color cues that would indicate spoilage, because consumers may not realize that meat has spoiled when its color remains bright red. Indeed, FDA itself has acknowledged consumers' reliance on color as a sign of freshness in expressing concerns about the use of carbon monoxide in tuna packaging, and the serious health risk posed when colorants mask the normal signs of spoilage.⁸⁴

While odor has been suggested as an alternative indicator of spoilage of meat packaged with carbon monoxide,⁸⁵ consumers obviously cannot detect the smell of packaged meat at the point of purchase to determine freshness. Even upon opening the package, however, consumers would not be able to rely upon odor, slime, or other organoleptic indicators of spoilage, because carbon dioxide-containing anaerobic packaging systems such as those that are the subject of the Pactiv and Precept GRAS notifications suppress the growth of aerobic spoilage organisms that produce these signals, while allowing other harmful yet imperceptible pathogens to flourish.⁸⁶ Indeed, even FDA has warned of this significant safety concern accompanying the

⁸⁴ See Letter from Diane E. Thompson, Associate Commissioner for Legislative Affairs, to Hon. Ray LaHood, Feb. 13, 1998 (Attachment 13), at 2 ("Consumers rely on the color of tuna to reflect its state of freshness. A process that inhibits the development of the telltale sensory changes that normally accompany decomposition or spoilage, such as the expected change in the color of the flesh, invite increased exposure to tuna products that are toxic, but not identifiable as such."). FDA ultimately issued a "no objection" letter in response to a GRAS notification for "tasteless smoke," of which carbon monoxide is a primary component, for use to "protect the taste, aroma, and color of seafood." See Agency Response Letter to GRAS Notice No. GRN 000015, available at <http://www.cfsan.fda.gov/~rdb/opa-g015.html>. However, that response has no relevance to the agency's consideration of the Pactiv and Precept GRAS notifications because section 173.350 of FDA regulations prohibits the use of carbon monoxide in the packaging of fresh meat products, due to the qualitatively distinct issues surrounding the use of colorants in fresh meat.

⁸⁵ See, e.g., Sørheim, O., *et al.*, "Technological, Hygienic and Toxicological Aspects of Carbon Monoxide Used in Modified-Atmosphere Packaging of Meat," 8 *Trends in Food Science & Technology* 307-312 (September 1997) (Attachment 14), at 311 ("A possible negative aspect of using CO in the MAP of retail meat is concern that consumers might misjudge the quality of a product, because its true microbiological status may be masked by its stable, cherry red carboxymyoglobin color. However, consumers will be able to detect spoilage by the presence of off-odours.") (citation omitted).

⁸⁶ See, e.g., Siliker, J.H. and Wolfe, S.K., "Microbiological Safety Considerations in Controlled-Atmosphere Storage of Meats," 34 *Food Technology* 59-63 (March 1980) (Attachment 15), at 59 (describing the fact that carbon dioxide in low-oxygen atmospheres "selectively inhibits the growth of Gram-negative bacteria, such as pseudomonads and other related psychrotrophs which grow rapidly and produce off-odors and -flavors in raw meats and poultry. . . . The organoleptic changes attended by the growth of lactic acid bacteria [in low-oxygen, elevated carbon dioxide packaging atmospheres] are less noticeable than those produced by the Gram-negative bacteria which develop upon meat in air atmospheres."); Farber, *supra* note 74, at 64 (explaining that the byproducts of the metabolism of the lactobacilli produced in anaerobic carbon dioxide-containing modified atmospheres "are inoffensive compared to the typical spoilage odors produced by the pseudomonads" that thrive in oxygenated atmospheres).

use of reduced oxygen packaging, cautioning that “the inhibition of the spoilage bacteria is significant because without these competing organisms, tell-tale signs signaling that the product is no longer fit for consumption will not occur.”⁸⁷ Of particular concern is the fact that consumers would not even be aware that they need to consider freshness criteria other than color or odor, such as “use by” date labeling, because fresh meat packaged with carbon monoxide is not required to be labeled as such, nor is the carbon monoxide’s coloring effect identified.⁸⁸ Accordingly, carbon monoxide in fresh meat packaging presents a serious public health risk because consumers will not be able to rely upon their accustomed indications of spoilage.⁸⁹

The European Commission’s Scientific Committee on Food squarely addressed these concerns about the likelihood that carbon monoxide will mask spoilage due to temperature abuse.⁹⁰ The Committee observed that “the inclusion of CO in MAP is controversial because the stable cherry-colour can last beyond the microbial shelf life of the meat and thus mask spoilage.”⁹¹ The extended shelf life attained by including carbon monoxide in packaging “may, therefore, under certain conditions imply increased risk of growth of pathogens.”⁹² The Committee concluded that carbon monoxide at levels of 0.3%-0.5% would be safe only if the temperature during storage and transport never exceeds 4°C (39°F), and observed in particular that some strains of *Salmonella* would grow at 10°C.⁹³ The Committee “wishes to point out that, should products be stored under inappropriate conditions, the presence of carbon monoxide may mask visual evidence of spoilage.”⁹⁴ In light of the Scientific Committee’s Opinion, the

⁸⁷ FDA Food Code, *supra* note 67, at 546.

⁸⁸ See Labuza and Fu, *supra* note 73, at 202 (stating that the recent trend to use MAP technology, “made with ‘invisible’ processing methods, which are not perceived as processing by the consumer, creates a new paradigm shift for food safety control” because of the potential to mask organoleptic signs of spoilage).

⁸⁹ See *id.* at 205 (“Sensory perceptions (e.g., meat color), evidence of metabolic by-products and types and levels of microorganisms are all valuable, and together give a full picture of food quality and safety.”).

⁹⁰ Opinion of the Scientific Committee on Food on the Use of Carbon Monoxide as Component of Packaging Gases in Modified Atmosphere Packaging for Fresh Meat, SCF/CS/ADD/MSAd/04 (December 18, 2001) (Attachment 16).

⁹¹ *Id.* at 4, citing Kropf, D.H., “Effect of Retail Display Conditions on Meat Color,” Proceedings of the 33rd Reciprocal Meat Conference, 15-32 (1980) (Attachment 17); see also Sørheim, O., *et al.*, “The Storage Life of Beef and Pork Packaged in an Atmosphere with Low Carbon Monoxide and High Carbon Dioxide,” 52 Meat Science 157-164 (1999) (Attachment 18), at 157 (“The inclusion of CO in MA for meat is controversial.”) and 163 (“An objection raised against using CO as a small component of a MA for retail-ready meat is the possibility that the colour stability can exceed the microbial shelf life, with the risk of masking spoilage of the meat.”) (citing Kropf, *supra*).

⁹² *Id.*, citing Nissen, *supra* note 78.

⁹³ *Id.* at 7.

⁹⁴ *Id.*

European Union refused to authorize carbon monoxide in fresh meat packaging, despite petitioning by the Norwegian government, precisely because of the dangerous effects in masking spoilage and encouraging consumer deception in ways that encourage consumption of unsafe meat.⁹⁵ As a result of the EU decision, the Norwegian meat industry was required to terminate the use of carbon monoxide in fresh meat packaging by June 2004, despite the history of commercial use in Norway since 1985. The EU has also banned carbon monoxide in fresh fish on the same grounds sustaining its ban in fresh meat products,⁹⁶ as have most other countries that have addressed the issue, including Canada, Japan, and Singapore.⁹⁷

Given FDA's recognition that home refrigerators can be expected to range between 5° and 10°C at best, in the hands of consumers, meat packaged with carbon monoxide will *never* be kept under the temperature conditions the Scientific Committee of the European Commission prescribed as necessary for safe use (at or below 4°C). Accordingly, under real world conditions, it is unavoidable that carbon monoxide in fresh meat will mask spoilage and promote consumer deception under the conditions of intended use.

The consumer safety risks from fresh meat packaged with carbon monoxide that has been exposed to temperature abuse are not ameliorated by "use by" date labeling such as that discussed in FDA's Agency Response Letter to GRN 143 and specified in FSIS Directive 7120.1 relating to use of carbon monoxide in accordance with that GRAS notification.⁹⁸ FDA has presented no consumer behavior evidence demonstrating that consumers would even consult date labeling where the color of the meat suggests freshness, and there is no means of enforcing consumer compliance with such labeling under real world conditions of use. More problematic is the fact that "use by" date labeling will likely amplify the public health risks by providing a false sense of security when the "use by" date has not passed and the meat still looks red, yet the meat has become spoiled due to microbial contamination resulting from temperature abuse. Notably, FSIS does not appear to have even considered the possibility of allowing the use of

⁹⁵ See EFTA Surveillance Authority Annual Report at 24 (2003), available at <http://www.eftasurv.int/information/annualreports/dbaFile4978.pdf> (relevant pages included as Attachment 19); Europarl News Report at 3 (June 11, 2003) (Attachment 20).

⁹⁶ See Letter from Jane M. Davies to Directors of Public Protection in Wales (August 9, 2004) (stating that carbon monoxide "causes an irreversible colour change in the fish flesh that has the potential to mislead consumers. As the product stays red even if it deteriorates or spoils, it is considered to be a potential public health hazard.") (Attachment 21).

⁹⁷ See, e.g., Julia Moskin, "Tuna's Red Glare? It Could Be Carbon Monoxide," N.Y. Times, Oct. 6, 2004 (Attachment 22); AVA Food Safety Awareness Programme Statement on Carbon Monoxide Treated Tuna, available at <http://www.ava.gov.sg/JAVASCRIPT/carbonMTuna.htm> (Attachment 23); Communiqué from Canadian Food Inspection Agency, Animal Products Directorate, Fish, Seafood and Production, to All Importers of Fish, regarding Fish Treated With Carbon Monoxide, June 17, 1999, available at <http://www.inspection.gc.ca/english/anima/fispoi/commun/19990617e.pdf> (Attachment 24).

⁹⁸ Products are required to be coded with a "Use or Freeze by" date not to exceed 28 days after packaging for ground meat and 35 days for whole muscle cuts.

sodium sulfite or paprika with “use by” labeling to ameliorate the deceptive coloring effects of these additives.

Finally, it cannot be said that cooking the meat will kill any pathogens and thereby counter any potential safety risks due to the presence of carbon monoxide in an oxygen-displacing modified atmosphere for fresh meat. *Clostridium botulinum* and *Clostridium perfringens*, which, if present, can thrive in such anaerobic atmospheres, are uniquely dangerous in fresh meat because their toxins are not destroyed by cooking. Even the aerobic pathogen *Salmonella* remains a serious food safety concern because many consumers fail to cook meat, and particularly ground beef, to interior temperatures sufficient to destroy this and other pathogens.⁹⁹

Given the record on consumer reliance upon meat color as an indicator of freshness, the inhibition of other organoleptic indicators of spoilage in modified atmosphere packaging, the documentation of extensive temperature abuse throughout the distribution and handling of fresh meat, and the inability of cooking to cure the harms of meat spoilage, FDA has pointed to no evidence demonstrating that no harm will result from carbon monoxide in fresh meat packaging under actual conditions of use. Without such evidence, carbon monoxide cannot be shown to be safe and suitable for use in fresh meat packaging, and therefore FDA cannot satisfy the statutory criteria at section 721(b)(1) for listing a color additive.

d. Carbon Monoxide in Fresh Meat Cannot Be Authorized Under FDCA Requirements for Food Additives and GRAS Substances

i Longstanding FDA Food Additive Regulations Prohibit Carbon Monoxide in Fresh Meat Packaging

FDA lacks authority to permit the use of carbon monoxide in fresh meat under FDCA requirements for food additives. Under well established FDA food additive regulations specifying the conditions in which carbon monoxide may be used to displace oxygen in food and beverage packaging, such use is expressly prohibited in fresh meat. Section 173.350 of FDA regulations prescribes the conditions under which “combustion product gas,” including carbon monoxide gas, can be used to displace oxygen in food packaging. The regulation specifies that such food packaging gases “may be safely used” in accordance with defined conditions, including controls to insure that gases “failing to meet the specifications . . . be prevented from reaching the food being treated.”¹⁰⁰ The rule authorizes the use of carbon monoxide gas in food packaging at levels up to 4.5 percent by volume, provided that “[i]t is used or intended for use to

⁹⁹ While USDA actively educates consumers that the only way to ensure that meats are cooked to safety is to use a food thermometer, only 2% of consumers report doing so. See FSIS, “The Food Safety Educator,” available at <http://www.fsis.usda.gov/OA/educator/educator3-4.htm>; see also Partnership for Food Safety Education, “Safe Cooking Fact Sheet,” available at http://www.fightbac.org/cook_facts.cfm.

¹⁰⁰ 21 C.F.R. 173.350(a).

displace or remove oxygen in . . . the packaging of beverage products and other food, except fresh meats" and other conditions are satisfied.¹⁰¹

In view of the breadth of the regulation, section 173.350 is properly construed as a food additive regulation that encompasses and regulates the conditions of use concerning carbon monoxide in food packaging to remove or displace oxygen. Particularly since the rule authorizes the use of carbon monoxide in all "beverage and other food" packaging, other than "fresh meat" products, it is unreasonable to construe the prohibition in "fresh meat" as a limitation on the scope of the food additive regulation, rather than a prescribed condition of use within the bounds of the rule. Clearly, where conditions of use have been established for a "food additive," these conditions of use cannot at the same time be excluded from the scope of food additive regulation as "GRAS." On this ground, the conditions of use of carbon monoxide in fresh meat packaging defined in the Pactiv and Precept GRAS notifications cannot qualify as "GRAS," as a matter of law, since these have already been established as "food additive" uses that are regulated directly and explicitly prohibited by section 173.350.

In addition, even if the "fresh meat" prohibition in 173.350 were construed as a limitation on the scope of authorized uses covered by the "food additive" regulation, such a limitation could be lawfully justified only on the grounds that carbon monoxide functions as a "color additive" in fresh meat, as opposed to other meat products for which carbon monoxide use is authorized under the FDA regulation. Given the breadth of the food additive regulation, and the authorization encompassing meat and poultry products other than "fresh meat" products, there would be no reasonable alternative basis for FDA to single out "fresh meat" for separate treatment. For example, there is no evidence suggesting that there are material differences in the toxicological safety of carbon monoxide that would support its use in all food and beverage products, including meat products, except for "fresh meat."

Moreover, in view of the restrictions on color additive uses in fresh meat historically, the "fresh meat" prohibition is justified by the particular hazard carbon monoxide presents under these conditions with respect to masking spoilage and deceptively encouraging consumption of unsafe meat. Such food safety and consumer deception issues are appropriately established as food additive specifications under FDCA section 409.¹⁰² While these same considerations bear on the status of carbon monoxide as a "color additive," as noted above, FDA is fully authorized to promulgate food additive specifications that complement the prohibited uses required under the color additive amendments.

FDA is not, however, authorized to pursue this radical departure from longstanding agency and FSIS policy prohibiting colorants in fresh meat via response to a GRAS notification. It is a well-established requirement of administrative law that where an agency

¹⁰¹ 21 C.F.R. 173.350(b), (c).

¹⁰² 21 U.S.C. 348.

departs from its prior positions, it must offer a reasoned explanation for its change in view.¹⁰³ Such agency action may be deemed arbitrary and capricious “if its rationale does not appear in the administrative record so that its decisionmaking path may reasonably be discerned.”¹⁰⁴ That record must demonstrate that the agency has considered all relevant factors.¹⁰⁵ Where FDA has drastically changed course after reviewing only the GRAS notifications of companies advocating the use of carbon monoxide in fresh meat packaging, the agency cannot demonstrate that it has met its burden of considering all relevant factors. Only after promulgating its new policy by means of notice and comment rulemaking on the public record, considering all relevant facts in its policy rationale could FDA comply with the applicable legal requirements.

ii Carbon Monoxide in Fresh Meat Cannot Satisfy FDCA Requirements for GRAS Substances

FDA lacks authority to condone the GRAS status of carbon monoxide in fresh meat packaging under the applicable FDCA requirements for GRAS substances. Under FDCA section 201(s), “GRAS” substances are distinguished from and excluded from the scope of the “food additive” definition.¹⁰⁶ Accordingly, under the same specified conditions of intended use, a substance cannot qualify at once as both a “GRAS” substance and a “food additive.” The FDCA makes clear that these are mutually exclusive categories, and establishes entirely separate and distinct regulatory requirements for food additives and GRAS substances. As discussed above, in 1962, FDA promulgated regulations making clear that the use of carbon monoxide to displace oxygen in food and beverage packaging is not GRAS, but rather must be regulated under the food additive provisions of the Act.¹⁰⁷ It is clear from the broad scope of the rule, which extends to all food and beverage packaging, including meat packaging, that the prohibition against carbon monoxide use in “fresh meat” packaging constitutes a specification within the scope of the food additive regulation, rather than a limitation upon the scope of the food additive rule itself. In short, there is no “fresh meat” gap in the scope of coverage of this food additive regulation which leaves room for any use of carbon monoxide in fresh meat packaging to evade premarket clearance requirements for food additives. Moreover, as discussed above, if anything, the fresh meat prohibition in section 173.350 is best explained as a reflection of the overlapping premarket clearance requirements for color additives. Since FDA has not listed carbon monoxide in fresh meat as required by the FDCA color additive provisions, FDA reasonably codified the prohibition as a specification in the relevant food additive regulation. Notably, GRAS status provides no insulation from FDA premarket clearance requirements for a color additive. In addition, since carbon monoxide is neither safe nor suitable for fresh meat

¹⁰³ *Department of the Navy v. FLRA*, 962 F.2d 48, 56 (D.C. Cir. 1992); *see also Hall v. McLaughlin*, 864 F.2d 868, 872 (D.C. Cir. 1989) (“[d]ivergence from agency precedent demands an explanation.”).

¹⁰⁴ *Chamber of Argentine-Paraguayan Producers of Quebracho Extract, et al. v. Holder*, 332 F. Supp. 2d 43, 49 (D.D.C. 2004).

¹⁰⁵ *Id.* at 48.

¹⁰⁶ 21 U.S.C. 321(s).

¹⁰⁷ 21 C.F.R. 173.350.

packaging because it promotes consumer deception, as discussed more fully at section B.4.c. of this petition, FDA is prohibited from approving such use under both the food additive or color additive provisions of the FDCA.¹⁰⁸

More fundamentally, despite the FDA responses to the Pactiv and Precept GRAS notifications, the sizable body of scientific evidence makes clear that the safety of carbon monoxide is not “generally recognized” as required by the FDCA. To the contrary, the safety of carbon monoxide in fresh meat has been widely challenged in the United States and internationally because of its capacity to mask spoilage and promote consumer deception.

Under FDA’s implementing regulations, the use of a food substance may be established as GRAS either through scientific procedures or, for a substance used in food before 1958, through experience based on common use in food. Under section 170.30(b), general recognition of safety through scientific procedures requires the same quantity and quality of scientific evidence as is required to obtain approval of the substance as a food additive and ordinarily is based upon published studies, which may be corroborated by unpublished studies and other data and information.¹⁰⁹ Under section 170.30(c) and 170.3(f), general recognition of safety through experience based on common use in foods requires a substantial history of consumption for food use by a significant number of consumers.¹¹⁰

For a substance to qualify as GRAS, there must be evidence that the substance is safe under the conditions of its intended use. FDA has defined “safe” as a reasonable certainty in the minds of competent scientists that the substance is not harmful under its intended conditions of use.¹¹¹ FDA has emphasized that a GRAS substance is distinguished from a food additive on the basis of the common knowledge about the safety of the substance for its intended use.¹¹² The “common knowledge” element of the GRAS standard includes two facets: “(1) the data and information relied on to establish the technical element must be generally available; and (2) there must be a basis to conclude that there is consensus among qualified experts about the safety of the substance for its intended use.”¹¹³ FDA advises that “an ongoing scientific discussion or

¹⁰⁸ 21 U.S.C. 348(c)(3) (prohibiting the approval of any food additive under conditions that “would promote deception of the consumer . . . or would otherwise result in adulteration or in misbranding of the food . . . ”); 21 U.S.C. 379e(b)(6) (prohibiting the listing of any color additive under conditions that “would promote deception of the consumer . . . or would otherwise result in misbranding or adulteration of the food . . . ”); 21 U.S.C. 342(b)(3)-(4) (defining adulterated food to include food in which “damage or inferiority has been concealed,” and food to which “any substance has been added . . . mak[ing] it appear better or of greater value than it is.”).

¹⁰⁹ 21 C.F.R. 170.30(b).

¹¹⁰ 21 C.F.R. 170.30(c) and 170.3(f).

¹¹¹ See FDA’s “Frequently Asked Questions About GRAS” (December 2004), available at <http://www.cfsan.fda.gov/~dms/grasguid.html>.

¹¹² 62 Fed. Reg. 18938 (April 17, 1997) (*see proposed* 170.36(c)(4)(i)(C)).

¹¹³ *Id.* at 18942.

controversy about safety concerns raised by available data would make it difficult to provide a basis for expert consensus about the safety of a substance for its intended use.”¹¹⁴

Plainly, the extensively documented controversy in the public literature about the safety of the use of carbon monoxide in fresh meat packaging belies any notion that the safety of this use is “generally recognized” among qualified experts.¹¹⁵ Moreover, a claim of GRAS status for this use of carbon monoxide cannot be maintained in light of the fact that the chemical is banned for use in meat or tuna packaging across much of the globe, including by Canada, the European Union, Japan and Singapore, because of the same food safety concerns outlined in this petition.¹¹⁶

Consideration of the relevant body of evidence makes clear that carbon monoxide in fresh meat is not GRAS, and there is substantial scientific evidence substantiating the serious nature of the food safety and consumer deception risks presented.

5. The FDCA and Implementing Regulations Require Label Declaration of the Use of Carbon Monoxide in Fresh Meat Packaging as a Fact Material to the Safe Handling of the Meat

Although, as detailed in this Petition, there are no grounds upon which FDA could lawfully allow the use of carbon monoxide in fresh meat packaging, even assuming *arguendo* that FDA had such authority, the agency would be required to implement FDCA labeling provisions requiring that the presence and purpose of the carbon monoxide in the packaging system be disclosed. The inclusion of carbon monoxide in modified atmosphere packaging of fresh meat is a fact material to the safe handling of the meat, and thus must be disclosed on the label in accordance with Sections 403(a) and 201(n) of the FDCA.

Section 403(a) states that a food shall be deemed misbranded if its labeling is false or misleading in any particular.¹¹⁷ Section 201(n) amplifies that provision by explaining that a food’s label is also misleading if it fails to reveal facts material in light of representations made, or material with respect to consequences which may result from the use of the food under customary or usual conditions of its use.¹¹⁸ The presence of carbon monoxide in fresh meat packaging is material under both prongs of section 201(n).

First, meat packaged in a carbon monoxide-containing modified atmosphere is represented as fresh and untreated. The use of carbon monoxide is a fact material in light of this representation and must be disclosed in labeling, because otherwise consumers will reasonably presume that the meat’s red color is a valid indication of its freshness and microbiological safety.

¹¹⁴ *Id.*

¹¹⁵ See discussion at section B.4.c.ii. of this petition, above, and accompanying footnotes.

¹¹⁶ See note 97, *supra*, and accompanying text.

¹¹⁷ 21 U.S.C. 343(a).

¹¹⁸ 21 U.S.C. 321(n).

Notably, FDA requires label declaration of the fact that a food has been irradiated¹¹⁹ – a process that produces material changes in the finished food that, in contradistinction to carbon monoxide use, serve to *increase* the safety of treated food. In the context of the food irradiation rulemaking, FDA indicated that, under sections 201(n) and 403(a), special labeling is required where a finished food is materially altered in a visually indiscernible way, and thus otherwise would be misrepresented as the traditional food.¹²⁰ In the case of irradiated food, the required labeling helps keep consumers from mistaking these foods for their traditional counterparts presenting significantly greater food safety risks. In the case of carbon monoxide-treated meats, the labeling that would be required by the FDCA would help keep consumers from mistaking these meats for traditionally packaged fresh meats, for the carbon monoxide-treated meats may give the appearance of freshness, but present significantly greater food safety risks.

The open date labeling that is a condition for use of the packaging system under GRN 143 does not obviate the materiality of the fact that carbon monoxide is present in fresh meat packaging. Consumers may disregard the “use or freeze by” date on the package if the meat still looks fresh. Even more concerning is the fact that meat may become spoiled before such date due to temperature abuse during distribution, but because the meat still looks fresh and shows no signs of spoilage and the date has not passed, consumers will reasonably assume that the meat is safe to consume.

Second, the use of carbon monoxide in fresh meat packaging is then also a fact material to the consequences that may result from the use of the meat under customary or usual conditions. Consumers accustomed to judging the freshness and safety of meat by color will likely store, prepare, and consume such meat as if it were as fresh as its color suggests, regardless of the actual age or safety of the meat. Bereft of the usual indicators that meat has spoiled, consumers could readily eat contaminated meat and suffer serious foodborne illness. Label disclosure of the presence and effect of carbon monoxide in fresh meat packaging is therefore required under sections 403(a) and 201(n), because a consumer who is not aware of the use of this chemical has no way of knowing that the appearance of the meat is not a reliable indicator that it is safe to consume.

Significantly, FDA required label declaration of the use of tasteless smoke, which includes carbon monoxide, in fresh tuna, where the smoke was used for purposes similar to that of carbon monoxide in fresh meat packaging, including to affect color.¹²¹ While the proponents of that GRAS notification positioned the tasteless smoke as a preservative, and therefore FDA required its label declaration under sections 403(k) and 403(i)(2) of the FDCA, the food safety considerations supporting disclosure apply with equal force to both uses of carbon monoxide-containing gases – the carbon monoxide affects the appearance of the meat or tuna in a manner

¹¹⁹ 21 C.F.R. 179.26(c).

¹²⁰ See 21 Fed. Reg. 13376, 13388 (April 18, 1986).

¹²¹ Letter from Janice F. Oliver, Deputy Director, CFSAN, to Martin J. Hahn, Hogan & Hartson (March 10, 2000) (“Agency Response Letter to GRAS Notice No. GRN 000015”), available at <http://www.cfsan.fda.gov/~rdb/opa-g015.html>.

that suggests freshness regardless of the actual age or safety of the food. Disclosure of the presence and effect of carbon monoxide, whether characterized as a chemical preservative or otherwise, is necessary to alert consumers to the fact that the appearance of the product is not a reliable indicator of its freshness or safety. It is precisely for this reason that color preservatives for use in fresh meat packaging must be identified on the label, and there is no justification for treating carbon monoxide differently.

Because the use of carbon monoxide in fresh meat packaging is a material fact in light of the representation that the meat is unprocessed and untreated and that its color is a reliable indicator of its freshness, and because of the serious food safety risks attendant to such representation, declaration of both the presence and the purpose of this use of carbon monoxide is required under sections 201(n) and 403(a) of the FDCA.

C. Environmental Impact

The action requested by this petition would result in the termination of FDA's responses to GRAS notifications and other actions preserving the status quo in conformance with well established law. The action requested is not expected to have a significant effect on the quality of the human environment, and is subject to categorical exclusion under 21 C.F.R. 25.30(h). To the best of Petitioner's knowledge, no extraordinary circumstances exist that would require an environmental assessment under 21 C.F.R. 25.21.

D. Economic Impact

Information on the economic impact of the action requested by this petition will be submitted if requested by the Commissioner.

E. Certification

The undersigned certify that, to the best of their knowledge and belief, this petition includes all information and views on which the petition relies, and that it includes representative data and information known to the petitioners which are unfavorable to the petition.

November 15, 2005

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For the foregoing reasons, this petition requests that FDA implement the actions requested to prohibit the use of carbon monoxide in fresh meat immediately.

Respectfully submitted,



Don Berdahl
Vice President/Lab Director
Kalsec, Inc.

cc: Dr. Andrew C. von Eschenbach, Acting Commissioner of Food and Drugs, FDA
Dr. Barbara J. Masters, Administrator, FSIS, USDA
Sheldon Bradshaw, Chief Counsel, FDA
Dr. Robert E. Brackett, Director, CFSAN, FDA
Dr. Laura M. Tarantino, Director, OFAS, FDA
Dr. Robert C. Post, Director, Labeling & Consumer Protection Staff, FSIS, USDA
Scott Gottlieb, Deputy Commissioner for Policy, FDA
Michael Landa, Deputy Director for Regulatory Affairs, CFSAN, FDA
Dr. Robert L. Martin, Deputy Division Director, OFAS, FDA
Dr. Rudolph Harris, Supervisor, OFAS, FDA
Dr. Robert L. Buchanan, Senior Science Advisor, CFSAN, FDA
Dr. Lane Highbarger, Consumer Safety Officer, OFAS, FDA
Dr. Bill Jones, Chemist, FSIS, USDA
Philip Derfler, Assistant Administrator, FSIS, USDA

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bolic pool, to be used again in protein synthesis or serve as a source of energy. Verdohemoglobin is reduced at the γ methene bridge to yield free bilirubin (orange-red, insoluble, and nonfilterable by the kidneys), which passes out of the reticuloendothelial cells into the plasma, where it is loosely bound to albumin. This bilirubin-albumin complex is carried to the liver, where it is conjugated with glucuronic acid within the liver cells. Normally most of this soluble bilirubin-diglucuronide passes into the biliary canaliculi. A small amount of this soluble, conjugated bilirubin is regurgitated back into the plasma, where it is again loosely attached to albumin. Because it is insoluble, it cannot be filtered readily by the kidneys.

From the biliary canaliculi, most of the soluble bilirubin-diglucuronide passes into the common bile duct and thence into the intestinal tract, where the bacterial flora removes the glucuronic acid, leaving the free bilirubin to be reduced in accordance with the type of bacterial flora present. One of the reduction products is urobilinogen, a colorless complex consisting mostly of stercobilinogen. When broad-spectrum antibiotics are given to patients, the bacterial flora is markedly diminished. This decreases the amount of bilirubin reduced to urobilinogen, and therefore urobilinogen excretion is reduced. Bilirubin thereby becomes the main bile pigment found in the feces. After the patient is taken off these antibiotics, the bacterial flora gradually returns, first reducing bilirubin to durobilinogen and then to stercobilinogen predominantly. A small amount of intestinal tract urobilinogen complex is reabsorbed and excreted again through the liver, or it appears in the urine. When feces and urine are exposed to air, they are oxidized to the urobilin group of compounds.

HEMOGLOBIN COMPOUNDS

The main function of hemoglobin in body metabolism is as a respiratory pigment in the form of oxyhemoglobin (scarlet red). As

erythrocytes flow along in single file in the delicate alveolar capillaries of the lung, the partial pressure (100 torr.) of oxygen in the alveolar air converts almost all the hemoglobin in these red blood cells to oxyhemoglobin, by a process of diffusion through the erythrocytic membrane. Because the association of oxygen and hemoglobin is loose and unstable, the oxygen readily diffuses back to the tissues for oxidative purposes, and the oxyhemoglobin then becomes reduced hemoglobin (dark red). There is, then, a concomitant release of base, which binds part of the incoming carbon dioxide. Carbon dioxide is also bound as a carbamate at the free amino groups of the hemoglobin molecule. A most important portion of the carbon dioxide diffuses from the plasma into the red blood cells, where catalysis by carbonic anhydrase joins it with water to form carbonic acid, which in turn dissociates into $(H)^+$ and $(HCO_3)^-$.

Carboxyhemoglobin is one of the abnormal hemoglobin pigments incapable of carrying oxygen. It is formed when hemoglobin in the red blood cells is exposed to carbon monoxide, which has an affinity 200 times greater for hemoglobin than oxygen does. When toxic amounts of carbon monoxide are present (from automobile exhaust fumes, for example), the blood is cherry red, and anoxia may result with subsequent death caused by irreversible tissue changes. Endogenous carbon monoxide production and subsequent respiratory excretion are related to heme degradation on a one-mole-to-one-mole basis. Since there is no other source of endogenous CO, measurement of its production rate accurately quantitates the catabolism of heme compounds, and thus also the rate of hemolysis.²³ Like oxyhemoglobin, carboxyhemoglobin is seen spectroscopically at 576 μ (Plate 8).

Methemoglobin is formed when hemoglobin in its deoxygenated state (reduced hemoglobin) is oxidized to the ferric form (iron normally exists in the ferrous state in the iron porphyrin complex of the heme portion of the hemoglobin molecule. See p.

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PART 121—FOOD ADDITIVES

Subpart D—Food Additives Permitted in Food for Human Consumption

COMBUSTION PRODUCT GAS

The Commissioner of Food and Drugs, having evaluated the data submitted by the Vitagen Corporation, 354 South Spring Street, Los Angeles 13, California, and other relevant material, has concluded that the following food additive regulation should issue with respect to the food additive combustion product gas used for the displacing and removal of oxygen in processing and packing of food. Therefore, pursuant to the provisions of the Federal Food, Drug, and Cosmetic Act (sec. 409(c)(1), 72 Stat. 1786; 21 U.S.C. 348(c)(1)), and under the authority delegated to the Commissioner by the Secretary of Health, Education, and Welfare (25 F.R. 8625), the food additive regulations (21 CFR 121) are amended by adding to Subpart D the following new section:

§ 121.1060 Combustion product gas.

The food additive combustion product gas may be safely used in the processing and packaging of the foods designated in paragraph (c) of this section for the purpose of removing and displacing oxygen in accordance with the following prescribed conditions:

(a) The food additive is manufactured by the controlled combustion in air of butane, propane, or natural gas. The combustion equipment shall be provided with an absorption-type filter capable of removing possible toxic impurities through which all gas used in the treatment of food shall pass; and with suitable controls to insure that any combustion products falling to meet the specifications provided in this section will be prevented from reaching the food being treated.

(b) The food additive meets the following specifications:

(1) Carbon monoxide content not to exceed 4.5 percent by volume.

(2) The ultraviolet absorbance in iso-octane solution in the range 255 millimicrons to 310 millimicrons not to exceed one-third of the standard reference absorbance when tested as described in paragraph (e) of this section.

(c) It is used or intended for use to displace or remove oxygen in the processing, storage, or packaging of citrus products, vegetable fats and vegetable oils, coffee, and wine.

(d) To assure safe use of the additive in addition to the other information required by the act, the label or labeling of the combustion device shall bear adequate directions for use to provide a combustion product gas that complies with the limitations prescribed in paragraph (b) of this section, including instructions to assure proper filtration.

(e) The food additive is tested for compliance with paragraph (b)(2) by the following empirical method:

Spectrophotometric measurements. All measurements are made in an ultraviolet spectrophotometer in optical cells of 5 centimeters in length, and in the range of 255 millimicrons to 310 millimicrons, under the same instrumental conditions. The standard reference absorbance is the absorbance at

large Room 540, 330 Independence Avenue SW., Washington 25, D.C., written objections thereto. Objections shall show wherein the person filing will be adversely affected by the order, and specify with particularity the provisions of the order deemed objectionable and the grounds for the objections. If a hearing is requested, the objections must state the issues for the hearing. A hearing will be granted if the objections are supported by grounds legally sufficient to justify the relief sought. Objections may be accompanied by a memorandum or brief in support thereof. All documents shall be filed in quantuplicate.

Effective date. This order shall be effective on the date of its publication in the FEDERAL REGISTER.

(Secs. 408(d)(2), 409(c)(4); 68 Stat. 512, 72 Stat. 1786; 21 U.S.C. 346a(d)(2), 348(c)(4))

Dated: July 26, 1961.

[SEAL] GEO. P. LARRICK,
Commissioner of Food and Drugs.

[F.R. Doc. 61-7270; Filed, Aug. 1, 1961;
8:50 a.m.]

PART 121—FOOD ADDITIVES

Subpart C—Food Additives Permitted in Animal Feed and Animal Feed Supplements

O,O-DIETHYL S-2-(ETHYLTHIO)ETHYL PHOSPHORODITHIOATE

Pursuant to sections 409 and 701 of the Federal Food, Drug, and Cosmetic Act and under the authority delegated to the Commissioner of Food and Drugs by the Secretary of Health, Education, and Welfare (25 F.R. 8625), § 121.215 of the food additive regulations (26 F.R. 2595) is revised to read as follows:

§ 121.215 O,O-Diethyl S-2-(ethylthio) ethyl phosphorodithioate.

A tolerance of 5 parts per million is established for residues of O,O-diethyl S-2-(ethylthio) ethyl phosphorodithioate, calculated as demeton, in dehydrated sugar beet pulp for livestock feed when present therein as a result of the application of the pesticide to the growing agricultural crop, provided that, if residues of demeton are also present, the total of both residues shall not exceed 5 parts per million.

This amendment does not require notice and public procedure since it is made for the purpose of bringing § 121.215 into conformity with the pesticide regulations.

Effective date. This order shall be effective on the date of its publication in the FEDERAL REGISTER.

(Secs. 409, 701; 52 Stat. 1055, 72 Stat. 1785; 21 U.S.C. 348, 371)

Dated: July 26, 1961.

[SEAL] GEO. P. LARRICK,
Commissioner of Food and Drugs.

[F.R. Doc. 61-7272; Filed, Aug. 1, 1961;
8:50 a.m.]

(S)-2-(ethylthio) ethyl phosphorothioates) on the same raw agricultural commodity, the total amount of such pesticides shall not yield more residue than that permitted by the larger of the two tolerances, calculated as demeton.

Section 120.105 is amended by adding thereto tolerances for residues of demeton in or on sugar beet tops and sugar beets. As amended § 120.105 reads as follows:

§ 120.105 Tolerances for residues of demeton.

Tolerances for residues of demeton (a mixture of O,O-diethyl O (and S)-2-(ethylthio) ethyl phosphorothioates) are established as follows:

12 parts per million in or on alfalfa hay, clover hay.

5 parts per million in or on almond hulls, fresh alfalfa, fresh clover, sugar beet tops.

1.25 parts per million in or on grapes, hops.

0.75 part per million in or on almonds, apples, apricots, broccoli, brussels sprouts, cabbage, cauliflower, celery, cottonseed, grapefruit, lemons, lettuce, muskmelons, oranges, peaches, pears, peas, pecans, peppers, plums (fresh prunes), potatoes, strawberries, tomatoes, walnuts.

0.5 part per million in or on sugar beets.

0.3 part per million in or on beans.

B. The Commissioner of Food and Drugs, having evaluated the data submitted in a petition filed by Chemagro Corporation, P.O. Box 4913, Kansas City 20, Missouri, and other relevant material, has concluded that the following regulation should issue with respect to residues of the food additive demeton present in dehydrated sugar beet pulp. Such residues have been shown to occur from application of the pesticide to sugar beets under agricultural uses provided for by a concurrent regulation under section 408 of the act. Therefore, pursuant to the provisions of the Federal Food, Drug, and Cosmetic Act (sec. 409(c)(4), 72 Stat. 1786; 21 U.S.C. 348(c)(4)), and under the authority delegated to the Commissioner by the Secretary of Health, Education, and Welfare (25 F.R. 8625), the food additive regulations (21 CFR Part 121) are amended by adding to Subpart C the following new section:

§ 121.221 Demeton.

A tolerance of 5 parts per million is established for residues of demeton (a mixture of O,O-diethyl O (and S)-2-(ethylthio) ethyl phosphorothioates) in dehydrated sugar beet pulp for livestock feed when present therein as a result of the application of the pesticide in the production of sugar beets, provided that if residues of O,O-diethyl S-2-(ethylthio) ethyl phosphorodithioate are also present, the total of both residues shall not exceed 5 parts per million.

Any person who will be adversely affected by the foregoing order may at any time prior to the thirtieth day from the date of its publication in the FEDERAL REGISTER file written objections with the Federal Register Clerk, Department of Health, Education, and Welfare, Washington 25, D.C., and

Particularly the provisions of the order deemed objectionable and the grounds for the objections. If a hearing is requested, the objections must state the issues for the hearing. A hearing will be granted if the objections are supported by grounds legally sufficient to justify the relief sought. Objections may be accompanied by a memorandum or brief in support thereof. All documents shall be filed in quintuplicate.

Effective date. This order shall be effective on the date of its publication in the FEDERAL REGISTER.

(Sec. 409(c)(1), 72 Stat. 1786; 21 U.S.C. 348(c)(1))

Dated: December 7, 1962.

GEO. P. LARRICK,
Commissioner of Food and Drugs.

(P.R. Doc. 62-12361; Filed, Dec. 13, 1962;
8:46 a.m.)

PART 121—FOOD ADDITIVES

Subpart D—Food Additives Permitted in Food for Human Consumption

COMBUSTION PRODUCT GAS

The Commissioner of Food and Drugs, having evaluated the data submitted in petitions filed by the Whirlpool Corporation, Benton Harbor, Michigan, and the Vitagen Corporation, 1263 Westwood Boulevard, Los Angeles, California, and other relevant material, has concluded that the food additive regulation with respect to combustion product gas should be amended as set forth below. Therefore, pursuant to the provisions of the Federal Food, Drug, and Cosmetic Act (Sec. 409(c)(1), 72 Stat. 1786; 21 U.S.C. 348(c)(1)), and under the authority delegated to the Commissioner by the Secretary of Health, Education, and Welfare (25 F.R. 8625), § 121.1060(c) (21 CFR 121.1060; 27 F.R. 4014) is amended to read as follows:

§ 121.1060 Combustion product gas.

(c) It is used or intended for use to displace or remove oxygen in the processing, storage, or packaging of beverage products and other food, except fresh meats.

(Any person who will be adversely affected by the foregoing order may at any time within 30 days from the date of its publication in the FEDERAL REGISTER file with the Hearing Clerk, Department of Health, Education, and Welfare, Room 5440, 330 Independence Avenue SW., Washington 25, D.C., written objections thereto. Objections shall show wherein the person filing will be adversely affected by the order and specify with particularity the provisions of the order deemed objectionable and the grounds for the objections. If a hearing is requested, the objections must state the issues for the hearing. A hearing will be granted if the objections are supported by grounds legally sufficient to justify the relief sought. Objections may be accompanied by a memorandum or brief in support thereof. All documents shall be filed in quintuplicate.

Effective date. This order shall be effective on the date of its publication in the FEDERAL REGISTER.

(Sec. 409(c)(1), 72 Stat. 1786; 21 U.S.C. 348(c)(1))

Dated: December 7, 1962.

GEO. P. LARRICK,
Commissioner of Food and Drugs.

(P.R. Doc. 62-12360; Filed, Dec. 13, 1962;
8:46 a.m.)

Title 39—POSTAL SERVICE

Chapter I—Post Office Department

PART 168—DIRECTORY OF INTERNATIONAL MAIL

Individual Country Amendments

The regulations of the Post Office Department in § 168.5 *Individual country regulations* are amended as follows:

I. In country "Bolivia", under Parcel Post, amend the item "Prohibitions" to read as follows:

Prohibitions. Firearms, daggers, black-jacks, brass knuckles, sidearms and concealable weapons.

Cigarette lighters.

Gambling devices.

Pharmaceutical and medicinal products, unless approved by the Bolivian health authorities. In case of doubt, senders should ascertain from the addressees in advance of mailing whether the medicine they desire to send will be admitted.

Articles which violate the Bolivian trademark laws.

Counterfeit or illegal currency; advertisements imitating currency or postage stamps, except for philatelic or numismatic catalogs.

Adulterated or harmful beverages or foodstuffs.

II. In country "Canada", as amended by 27 F.R. 404, 27 F.R. 10369, under Parcel Post, the item "Prohibitions" is amended by revising the sixth paragraph to include "Plumage and skins of wild birds" and by adding a new paragraph at the end thereof to prescribe regulations for importing meat. As so amended, paragraph six and the new paragraph read as follows:

Prohibitions. . . .

Commercial tags of metal. Prison-made goods being sold or intended for sale by a person or firm. Plumage and skins of wild birds.

Meat and meat food products, unless federally inspected and passed and marked accordingly. If intended for sale, export certification by the United States Department of Agriculture is also required. Meat or meat food product for personal use is exempt from export certification, but the addressee is required to certify to the Canadian authorities that it will not be offered for sale in Canada.

III. In country "Japan", under Parcel Post, the item "Prohibitions" is amended by revising the second paragraph to in-

clude wool samples among animal products. As so amended, the second paragraph reads as follows:

Prohibitions. . . .

The following must be accompanied by official inspection certificates showing that they are free from domestic animals' infectious disease: Meat, bones, skin, hair, feathers, horns or hoofs of hoofed animals, rabbits, or poultry, wool samples, poultry eggs for hatching; honey bees.

IV. In country "Kenya and Uganda", as amended by 27 F.R. 3738, 27 F.R. 5659, under Parcel Post, amend the tabular information immediately following the item "Air parcel rates" by striking out "Weight limit: 11 pounds", and inserting in lieu thereof "Weight limit: 22 pounds."

V. In country "Laos", as amended by 27 F.R. 8592, amend the item "Observations" where it appears both under Postal Union Mail and Parcel Post, to respectively read as follows:

Observations. The following are the only post offices in operation:

Vientiane.	Paksé.
Honetai.	Paksong.
Luangprabang.	Khongsedone.
Sayaboury.	Champassak.
Paksane.	Muong Kong.
Khammouane.	Saravane.
Savannakhet.	Attapeu.

Observations. See the item "Observations" under Postal Union Mail for post offices which are in operation.

VI. In country "Tanganyika Territory" under Parcel Post, amend the tabular information immediately following the item "Air parcel rates" by striking out "Weight limits: 11 pounds" and inserting in lieu thereof "Weight limits: 22 pounds".

VII. In country "Thailand", as amended by 27 F.R. 7022, under Parcel Post, make the following changes to show that insured parcel post service is available.

A. Amend the tabular information immediately following the item "Air parcel rates" to read as follows:

Weight limit: 22 pounds
Sealing: Insured parcels must, and ordinary parcels may be sealed
Registration: No
Insurance: Yes
Postal forms required:
1 Form 2922
1 Form 2966

B. Strike out the item "Indemnity. No provision." and insert in lieu thereof the following:

Insurance. The following insurance fees and limits of indemnity apply:

Limit of indemnity:	Fee, cents
Not over \$10-----	20
From \$10.01 to \$25-----	25
From \$25.01 to \$50-----	35
From \$50.01 to \$100-----	55

Insured parcels may only be addressed to Bangkok or Dhonburi.

Print on the wrapper, near the "INSURED" endorsement and number, the amount for which the parcel is insured. This amount shall be shown in United

§ 173.342

(b) They are added in an amount not in excess of that reasonably required to inhibit foaming.

[42 FR 14526, Mar. 15, 1977, as amended at 43 FR 2872, Jan. 20, 1978; 46 FR 30493, June 9, 1981; 46 FR 57476, Nov. 24, 1981; 60 FR 54036, Oct. 19, 1995; 61 FR 632, Jan. 9, 1996; 63 FR 29134, May 28, 1998]

§ 173.342 Chlorofluorocarbon 113 and perfluorohexane.

A mixture of 99 percent chlorofluorocarbon 113 (1,1,2-trichloro-1,2,2-trifluoroethane) (CAS Reg. No. 76-13-1, also known as fluorocarbon 113, CFC 113 and FC 113) and 1 percent perfluorohexane (CAS Reg. No. 355-42-0) may be safely used in accordance with the following prescribed conditions:

(a) The additive chlorofluorocarbon 113 has a purity of not less than 99.99 percent.

(b) The additive mixture is intended for use to quickly cool or crust-freeze chickens sealed in intact bags composed of substances regulated in parts 174, 175, 177, 178, and § 179.45 of this chapter and conforming to any limitations or specifications in such regulations.

[55 FR 8913, Mar. 9, 1990]

§ 173.345 Chloropentafluoroethane.

The food additive chloropentafluoroethane may be safely used in food in accordance with the following prescribed conditions:

(a) The food additive has a purity of not less than 99.97 percent, and contains not more than 200 parts per million saturated fluoro compounds and 10 parts per million unsaturated fluoro compounds as impurities.

(b) The additive is used or intended for use alone or with one or more of the following substances: Carbon dioxide, nitrous oxide, propane, and octafluorocyclobutane complying with § 173.360, as an aerating agent for foamed or sprayed food products, with any propellant effect being incidental and no more than is minimally necessary to achieve the aerating function, except that use is not permitted for those standardized foods that do not provide for such use.

(c) To assure safe use of the additive

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(1) The label of the food additive container shall bear, in addition to the other information required by the act, the following:

(i) The name of the additive, chloropentafluoroethane.

(ii) The percentage of the additive present in the case of a mixture.

(iii) The designation "food grade".

(2) The label or labeling of the food additive container shall bear adequate directions for use.

[42 FR 14526, Mar. 15, 1977, as amended at 43 FR 11317, Mar. 17, 1978; 43 FR 14644, Apr. 7, 1978]

§ 173.350 Combustion product gas.

The food additive combustion product gas may be safely used in the processing and packaging of the foods designated in paragraph (c) of this section for the purpose of removing and displacing oxygen in accordance with the following prescribed conditions:

(a) The food additive is manufactured by the controlled combustion in air of butane, propane, or natural gas. The combustion equipment shall be provided with an absorption-type filter capable of removing possible toxic impurities, through which all gas used in the treatment of food shall pass; and with suitable controls to insure that any combustion products failing to meet the specifications provided in this section will be prevented from reaching the food being treated.

(b) The food additive meets the following specifications:

(1) Carbon monoxide content not to exceed 4.5 percent by volume.

(2) The ultraviolet absorbance in isooctane solution in the range 255 millimicrons to 310 millimicrons not to exceed one-third of the standard reference absorbance when tested as described in paragraph (e) of this section.

(c) It is used or intended for use to displace or remove oxygen in the processing, storage, or packaging of beverage products and other food, except fresh meats.

(d) To assure safe use of the additive in addition to the other information required by the act, the label or labeling of the combustion device shall bear adequate directions for use to provide a combustion product gas that complies

with the limitations prescribed in paragraph (b) of this section, including instructions to assure proper filtration.

(e) The food additive is tested for compliance with paragraph (b)(2) by the following empirical method:

Spectrophotometric measurements. All measurements are made in an ultraviolet spectrophotometer in optical cells of 5 centimeters in length, and in the range of 255 millimicrons to 310 millimicrons, under the same instrumental conditions. The standard reference absorbance is the absorbance at 275 millimicrons of a standard reference solution of naphthalene (National Bureau of Standards Material No. 577 or equivalent in purity) containing a concentration of 1.4 milligrams per liter in purified isooctane, measured against isooctane of the same spectral purity in 5-centimeter cells. (This absorbance will be approximately 0.30.)

Solvent. The solvent used is pure grade isooctane having an ultraviolet absorbance not to exceed 0.05 measured against distilled water as a reference. Upon passage of purified inert gas through some isooctane under the identical conditions of the test, a lowering of the absorbance value has been observed. The absorbance of isooctane to be used in this procedure shall not be more than 0.02 lower in the range 255 millimicrons to 310 millimicrons, inclusive, than that of the untreated solvent as measured in a 5-centimeter cell. If necessary to obtain the prescribed purities, the isooctane may be passed through activated silica gel.

Apparatus. To assure reproducible results, the additive is passed into the isooctane solution through a gas-absorption train consisting of the following components and necessary connections:

1. A gas flow meter with a range up to 30 liters per hour provided with a constant differential relay or other device to maintain a constant flow rate independent of the input pressure.

2. An absorption apparatus consisting of an inlet gas dispersion tube inserted to the bottom of a covered cylindrical vessel with a suitable outlet on the vessel for effluent gas. The dimensions and arrangement of tube and vessel are such that the inlet tube introduces the gas at a point not above 5/4 inches below the surface of the solvent through a sintered glass outlet. The dimensions of the vessel are such, and both inlet and vessel are so designed, that the gas can be bubbled through 60 milliliters of isooctane solvent at a rate up to 30 liters per hour without mechanical loss of solvent. The level corresponding to 60 milliliters should be marked on the vessel.

3. A cooling bath containing crushed ice and water to permit immersion of the absorption vessel at least to the solvent level mark.

Caution. The various parts of the absorption train must be connected by gas-tight tubing and joints composed of materials which will neither remove components from nor add components to the gas stream. The gas source is connected in series to the flow-rate device, the flow meter, and the absorption apparatus in that order. Ventilation should be provided for the effluent gases which may contain carbon monoxide.

Sampling procedure. Immerse the gas-absorption apparatus containing 60 milliliters of isooctane in the coolant bath so that the solvent is completely immersed. Cool for at least 15 minutes and then pass 120 liters of the test gas through the absorption train at a rate of 30 liters per hour or less. Maintain the coolant bath at 0 °C throughout. Remove the absorption vessel from the bath, disconnect, and warm to room temperature. Add isooctane to bring the contents of the absorption vessel to 60 milliliters, and mix. Determine the absorbance of the solution in the 5-centimeter cell in the range 255 millimicrons to 310 millimicrons, inclusive, compared to isooctane. The absorbance of the solution of combustion product gas shall not exceed that of the isooctane solvent at any wavelength in the specified range by more than one-third of the standard reference absorbance.

§ 173.355 Dichlorodifluoromethane.

The food additive dichlorodifluoromethane may be safely used in food in accordance with the following prescribed conditions:

- (a) The additive has a purity of not less than 99.97 percent.

- (b) It is used or intended for use, in accordance with good manufacturing practice, as a direct-contact freezing agent for foods.

- (c) To assure safe use of the additive:

- (1) The label of its container shall bear, in addition to the other information required by the act, the following:

- (i) The name of the additive, dichlorodifluoromethane, with or without the parenthetical name "Food Freezant 12".

- (ii) The designation "food grade".

- (2) The label or labeling of the food additive container shall bear adequate directions for use.

§ 173.357 Materials used as fixing agents in the immobilization of enzyme preparations.

Fixing agents may be safely used in the immobilization of enzyme preparations in accordance with the following conditions:

§ 173.350 Combustion product gas.

The food additive combustion product gas may be safely used in the processing and packaging of the foods designated in paragraph (c) of this section for the purpose of removing and displacing oxygen in accordance with the following prescribed conditions:

(a) The food additive is manufactured by the controlled combustion in air of butane, propane, or natural gas. The combustion equipment shall be provided with an absorption-type filter capable of removing possible toxic impurities, through which all gas used in the treatment of food shall pass; and with suitable controls to insure that any combustion products failing to meet the specifications provided in this section will be prevented from reaching the food being treated.

(b) The food additive meets the following specifications:

(1) Carbon monoxide content not to exceed 4.5 percent by volume.

(2) The ultraviolet absorbance in isooctane solution in the range 255 millimicrons to 310 millimicrons not to exceed one-third of the standard reference absorbance when tested as described in paragraph (e) of this section.

(c) It is used or intended for use to displace or remove oxygen in the processing, storage, or packaging of beverage products and other food, except fresh meats.

(d) To assure safe use of the additive in addition to the other information required by the act, the label or labeling of the combustion device shall bear adequate directions for use to provide a combustion product gas that complies with the limitations prescribed in paragraph (b) of this section, including instructions to assure proper filtration.

(e) The food additive is tested for compliance with paragraph (b)(2) by the following empirical method:

Spectrophotometric measurements. All measurements are made in an ultraviolet spectrophotometer in optical cells of 5 centimeters in length, and in the range of 255 millimicrons to 310 millimicrons, under the same instrumental conditions. The standard reference absorbance is the absorbance at 275 millimicrons of a standard reference solution of naphthalene (National Bureau of Standards Material No. 577 or equivalent in purity) containing a concentration of 1.4 milligrams per liter in purified isooctane, measured

against isooctane of the same spectral purity in 5-centimeter cells. (This absorbance will be approximately 0.30.)

Solvent. The solvent used is pure grade isooctane having an ultraviolet absorbance not to exceed 0.05 measured against distilled water as a reference. Upon passage of purified inert gas through some isooctane under the identical conditions of the test, a lowering of the absorbance value has been observed. The absorbance of isooctane to be used in this procedure shall not be more than 0.02 lower in the range 255 millimicrons to 310 millimicrons, inclusive, than that of the untreated solvent as measured in a 5-centimeter cell. If necessary to obtain the prescribed purities, the isooctane may be passed through activated silica gel.

Apparatus. To assure reproducible results, the additive is passed into the isooctane solution through a gas-absorption train consisting of the following components and necessary connections:

1. A gas flow meter with a range up to 30 liters per hour provided with a constant differential relay or other device to maintain a constant flow rate independent of the input pressure.

2. An absorption apparatus consisting of an inlet gas dispersion tube inserted to the bottom of a covered cylindrical vessel with a suitable outlet on the vessel for effluent gas. The dimensions and arrangement of tube and vessel are such that the inlet tube introduces the gas at a point not above 5/4 inches below the surface of the solvent through a sintered glass outlet. The dimensions of the vessel are such, and both inlet and vessel are so designed, that the gas can be bubbled through 60 milliliters of isooctane solvent at a rate up to 30 liters per hour without mechanical loss of solvent. The level corresponding to 60 milliliters should be marked on the vessel.

3. A cooling bath containing crushed ice and water to permit immersion of the absorption vessel at least to the solvent level mark.

Caution. The various parts of the absorption train must be connected by gas-tight tubing and joints composed of materials which will neither remove components from nor add components to the gas stream. The gas source is connected in series to the flow-rate device, the flow meter, and the absorption apparatus in that order. Ventilation should be provided for the effluent gases which may contain carbon monoxide.

Sampling procedure. Immerse the gas-absorption apparatus containing 60 milliliters of isooctane in the coolant bath so that the solvent is completely immersed. Cool for at least 15 minutes and then pass 120 liters of the test gas through the absorption train at a rate of 30 liters per hour or less. Maintain the coolant bath at 0 °C throughout. Remove the absorption vessel from the bath, disconnect, and warm to room temperature.

§ 173.355

21 CFR Ch. I (4-1-04 Edition)

Add isooctane to bring the contents of the absorption vessel to 60 milliliters, and mix. Determine the absorbance of the solution in the 5-centimeter cell in the range 255 millimicrons to 310 millimicrons, inclusive, compared to isooctane. The absorbance of the solution of combustion product gas shall not exceed that of the isooctane solvent at any wavelength in the specified range by more than one-third of the standard reference absorbance.

§ 173.355 Dichlorodifluoromethane.

The food additive dichlorodifluoromethane may be safely used in food in accordance with the following prescribed conditions:

(a) The additive has a purity of not less than 99.97 percent.

(b) It is used or intended for use, in accordance with good manufacturing practice, as a direct-contact freezing agent for foods.

(c) To assure safe use of the additive:

(1) The label of its container shall bear, in addition to the other information required by the act, the following:

(i) The name of the additive, dichlorodifluoromethane, with or without the parenthetical name "Food Freezant 12".

(ii) The designation "food grade".

(2) The label or labeling of the food additive container shall bear adequate directions for use.

§ 173.357 Materials used as fixing agents in the immobilization of enzyme preparations.

Fixing agents may be safely used in the immobilization of enzyme preparations in accordance with the following conditions:

(a) The materials consist of one or more of the following:

(1) Substances generally recognized as safe in food.

(2) Substances identified in this subparagraph and subject to such limitations as are provided:

Substances	Limitations
Acrylamide-acrylic acid resin: Complying with § 173.5(a)(1) and (b) of this chapter.	May be used as a fixing material in the immobilization of glucose isomerase enzyme preparations for use in the manufacture of high fructose corn syrup, in accordance with § 184.1372 of this chapter.
Cellulose triacetate	May be used as a fixing material in the immobilization of lactase for use in reducing the lactose content of milk.
Diethylaminoethyl-cellulose	May be used as a fixing material in the immobilization of glucose isomerase enzyme preparations for use in the manufacture of high fructose corn syrup, in accordance with § 184.1372 of this chapter.
Dimethylamine-epichlorohydrin resin: Complying with § 173.60(a) and (b) of this chapter.	May be used as a fixing material in the immobilization of glucose isomerase enzyme preparations for use in the manufacture of high fructose corn syrup, in accordance with § 184.1372 of this chapter.
Glutaraldehyde	Do.
Periodic acid (CAS Reg. No. 10450-60-9) ..	



DEPARTMENT OF HEALTH & HUMAN SERVICES

Public Health Service

Food and Drug Administration
Washington, DC 20204

Eric Greenberg
Ungaretti and Harris
3500 Three First National Plaza
Chicago, IL, 60602-4405

Re: GRAS Notice No. GRN 000083

Dear Mr. Greenberg:

The Food and Drug Administration (FDA) is responding to the notice, dated August 29, 2001, that Ungaretti and Harris submitted on behalf of Pactiv Corporation (Pactiv) in accordance with the agency's proposed regulation, proposed 21 CFR 170.36 (62 FR 18938; April 17, 1997; Substances Generally Recognized as Safe (GRAS)). FDA received the notice on September 4, 2001, and designated it as GRAS Notice No. GRN 000083.

The subject of the notice is carbon monoxide (CO). The notice informs FDA of the view of Pactiv Corporation (Pactiv) that CO is GRAS, through scientific procedures, for use as a component of a gas mixture in a modified atmosphere packaging (MAP) system. The level of CO in this MAP system is 0.4 percent. The other components of the MAP system are carbon dioxide (30 percent) and nitrogen (69.6 percent). The MAP system would be used for packaging fresh cuts of case ready muscle meat and ground case ready meat to maintain wholesomeness, provide flexibility in distribution, and reduce shrinkage of the meat. The case ready meats would be removed from the MAP system prior to retail display.

As part of its notice, Pactiv includes letters from a panel of individuals (Pactiv's GRAS panel) who evaluated the data and information that are the basis for Pactiv's GRAS determination. Pactiv considers the members of its GRAS panel to be qualified by scientific training and experience to evaluate the safety of substances added to food. Pactiv's GRAS panel evaluated information and data on the chemical identity, manufacture and processing, conditions of proposed use, and estimated daily intakes of CO used in a MAP system for meat. Pactiv's GRAS panel also evaluated studies (published and unpublished) of the effects of CO used in a MAP system for meat. Members of the GRAS panel reviewed and evaluated the publicly available information summarized in the GRAS notice. Based on the data and information reviewed, Pactiv's GRAS panel concludes that CO, when produced in accordance with current good manufacturing practice and meeting appropriate food grade specifications, is GRAS, through scientific procedures under the conditions of its intended use.

The notice describes publicly available information pertaining to the identity and characteristic properties of CO. Carbon monoxide (Chemical Abstracts Service Registry Number 630-08-0) is a colorless, odorless, gas. The notice includes a list of properties of CO and identifies the

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manufacturer who currently supplies CO to Pactiv. Pactiv intends to use CO at a minimum purity of 99.99 percent ("commercial grade"). Pactiv includes a list of specifications for CO with limits on the levels of other gases and considers CO of this purity to be "food grade."

The notice describes information about existing regulations and notices regarding food substances that contain CO as a significant component:

- Wood smoke, which includes CO as a component, is permitted by regulation as an ingredient in meat and poultry products under regulations issued by the U.S. Department of Agriculture (9 CFR 318.7(c)(4), 381.147(c)(4) and 424.21(c)).
- Combustion product gas, which includes CO as a component at a maximum level of 4.5 percent by volume, is approved for use in the production of beverages and other foods (except fresh meat) under FDA's regulations (21 CFR 173.350).
- Tasteless smoke, which includes CO as a primary component, is the subject of GRN 000015 for use on raw tuna, before it is frozen, to preserve its taste, aroma, texture, and color. In response to GRN 000015, FDA had no questions regarding the notifier's conclusion that tasteless smoke is GRAS under the intended conditions of use.

The notice describes the estimated consumption of CO per meal as a consequence of its intended use as a component in a MAP system for storing meat. Assuming that 30 percent of the CO present in the MAP is absorbed into the meat and that there is an 85 percent reduction of CO due to cooking the meat, Pactiv calculates a realistic intake estimate to be 0.084 milligrams (mg) CO per meal. Pactiv also calculates a worst case intake estimate to be 1.88 mg CO per meal, assuming that 100 percent of the CO present in the MAP is absorbed into the meat and that there is no reduction in CO during cooking. Pactiv cites published articles to support the assumptions used in the realistic exposure estimate and to support the conclusion that exposure to CO is safe at this level.

The notice describes published reports of studies demonstrating the technical effect and safety of using CO as a component of a MAP system (similar to the MAP system that is the subject of GRN 000083) for storing meat. These reports include published data (microbial growth profiles and odor and color data) from meat stored in MAP containing CO, CO₂, and N₂, and meat stored in MAP containing only CO₂ and N₂. Pactiv concludes that the presence of CO in MAP systems allows the meat to maintain a desirable red color during storage. In addition, CO neither affects the ability of the MAP system to slow the growth of a variety of microorganisms, nor affects the characteristic odor of meat spoilage.

The notice describes an unpublished study using the MAP system that is the subject of GRN 000083. The study examined the effects of the system on initial meat color, stability of color during display, and the relationship between color deterioration and microbial growth. The notice also includes unpublished pictures that compare the ageing (color deterioration) of meats stored for 20 days in an environment of CO, CO₂, and N₂, to the ageing of fresh cut meat and the ageing of meat stored in a high oxygen environment. From these data, Pactiv concludes that once meat is removed from a MAP system containing CO, its color deteriorates at a similar rate to

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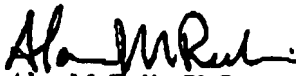
that of meat that has not been exposed to CO. Pactiv also concludes that the use of CO in a MAP system does not result in red color life extension that could mask microbial spoilage of the meat.

Based on the information provided by Pactiv, as well as other information available to FDA, the agency has no questions at this time regarding Pactiv's conclusion that CO is GRAS under the intended conditions of use. The agency has not, however, made its own determination regarding the GRAS status of the subject use of CO. As always, it is the continuing responsibility of Pactiv to ensure that food ingredients that the firm markets are safe, and are otherwise in compliance with all applicable legal and regulatory requirements.

During its evaluation of GRN 000083, OFAS consulted with the Labeling and Consumer Protection Staff of the Food Safety and Inspection Service (FSIS) of the United States Department of Agriculture regarding the use of CO in meat products. Based on the information submitted by Pactiv, FSIS has concluded that the MAP system (ActiveTech™ 2001) as described in Pactiv's notice, and used under the conditions stated in Pactiv's notice, would be acceptable for packaging red meat cuts and ground meat. In FSIS' view, Pactiv has demonstrated that this MAP system complies with FDA's definition of a processing aid that appears in labeling regulations (21 CFR 101.100(a)(3)). There is no lasting functional effect in the food and there is an insignificant amount of carbon monoxide present in the finished product under the proposed conditions of use. As such, similar to uses of other MAP gases (e.g., nitrogen), there are no labeling issues in regard to meat cuts and ground meat packaged using this MAP. Additionally, when considering the use of a food ingredient or additive in a meat product, FSIS historically has treated each livestock species separately. However, in this case, the data submitted by Pactiv can be extrapolated to all species of livestock. If you have any additional questions, you should direct your inquiry to Dr. Robert Post, Director, Labeling and Consumer Protection Staff, Office of Policy, Program Development and Evaluation, Food Safety and Inspection Service, 300 12th Street, SW, Room 602, Washington, DC 20250-3700. The telephone number of his office is (202) 205-0279 and the FAX number is (202) 205-3625.

In accordance with proposed 21 CFR 170.36(f), a copy of the text of this letter, as well as a copy of the information in your notice that conforms to the information in proposed 21 CFR 170.36(c)(1), is available for public review and copying on the homepage of the Office of Food Additive Safety (on the Internet at <http://www.cfsan.fda.gov/~lrd/foodadd.html>).

Sincerely,



Alan M. Rulis, Ph.D.

Director
Office of Food Additive Safety
Center for Food Safety
and Applied Nutrition